Addressing Our Country's Need for Biofuels:

"Looking at chemical engineers establishing a bio-based economy in the Visayas region"

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2nd PIChE VISAYAS REGIONAL CONFERENCE

BIOMASS B&B BIOFUEL

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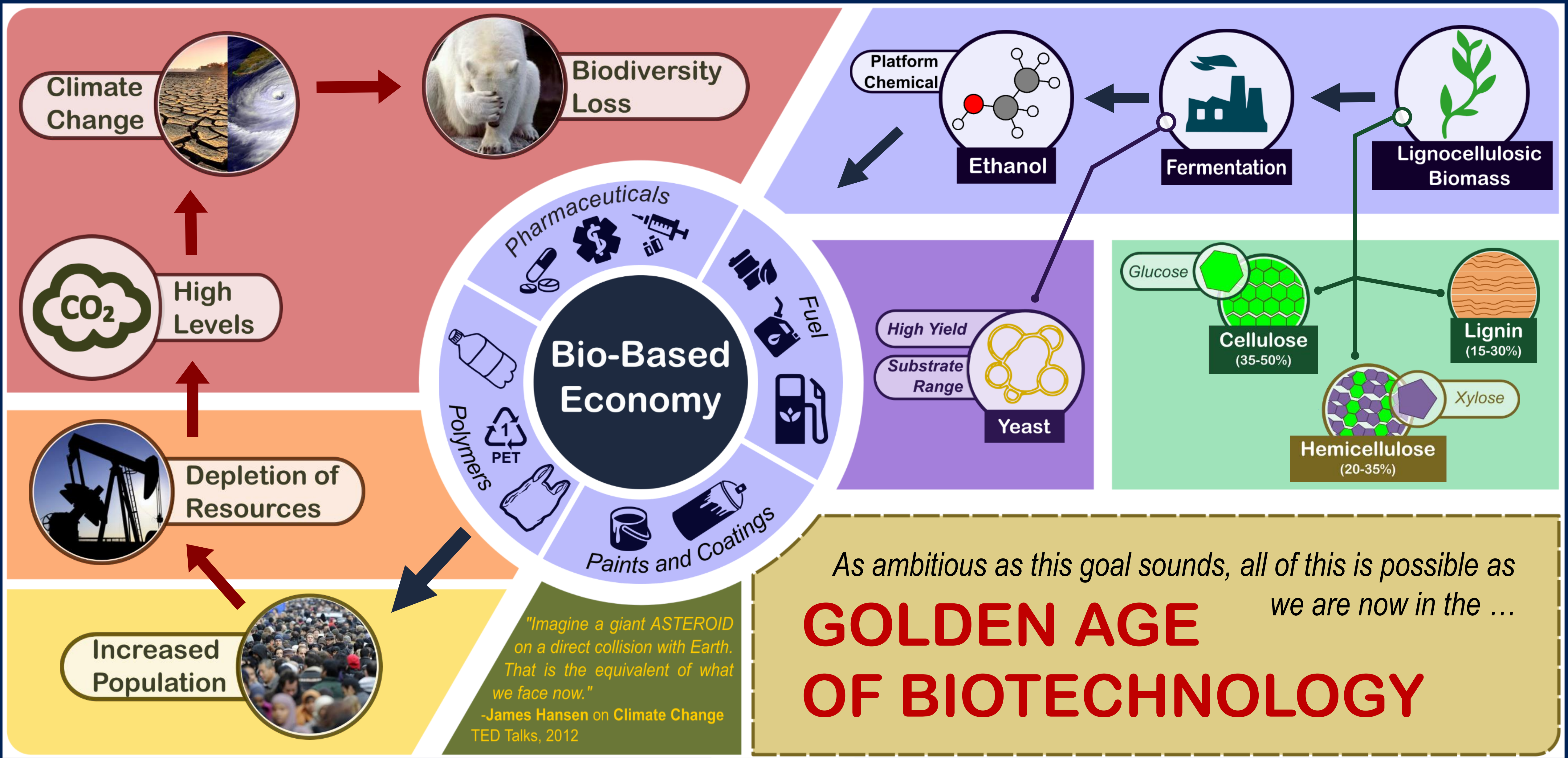
Looking at one of the BIGGEST problems that the world is facing today…

Climate Change

"Climate change is moving faster than we are … We must listen to the Earth's best scientists."

-António Guterres
UN Secretary General
Adapted from his 2018 UN General Assembly address

ENGINEERING YEAST: A QUEST AGAINST CLIMATE CHANGE



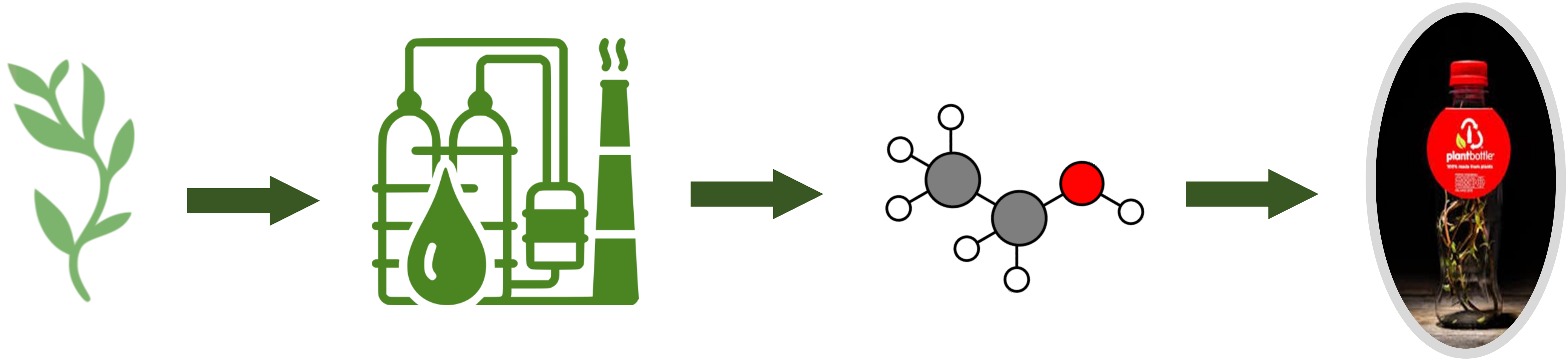
IAN DOMINIC FLORMATA TABAÑAG

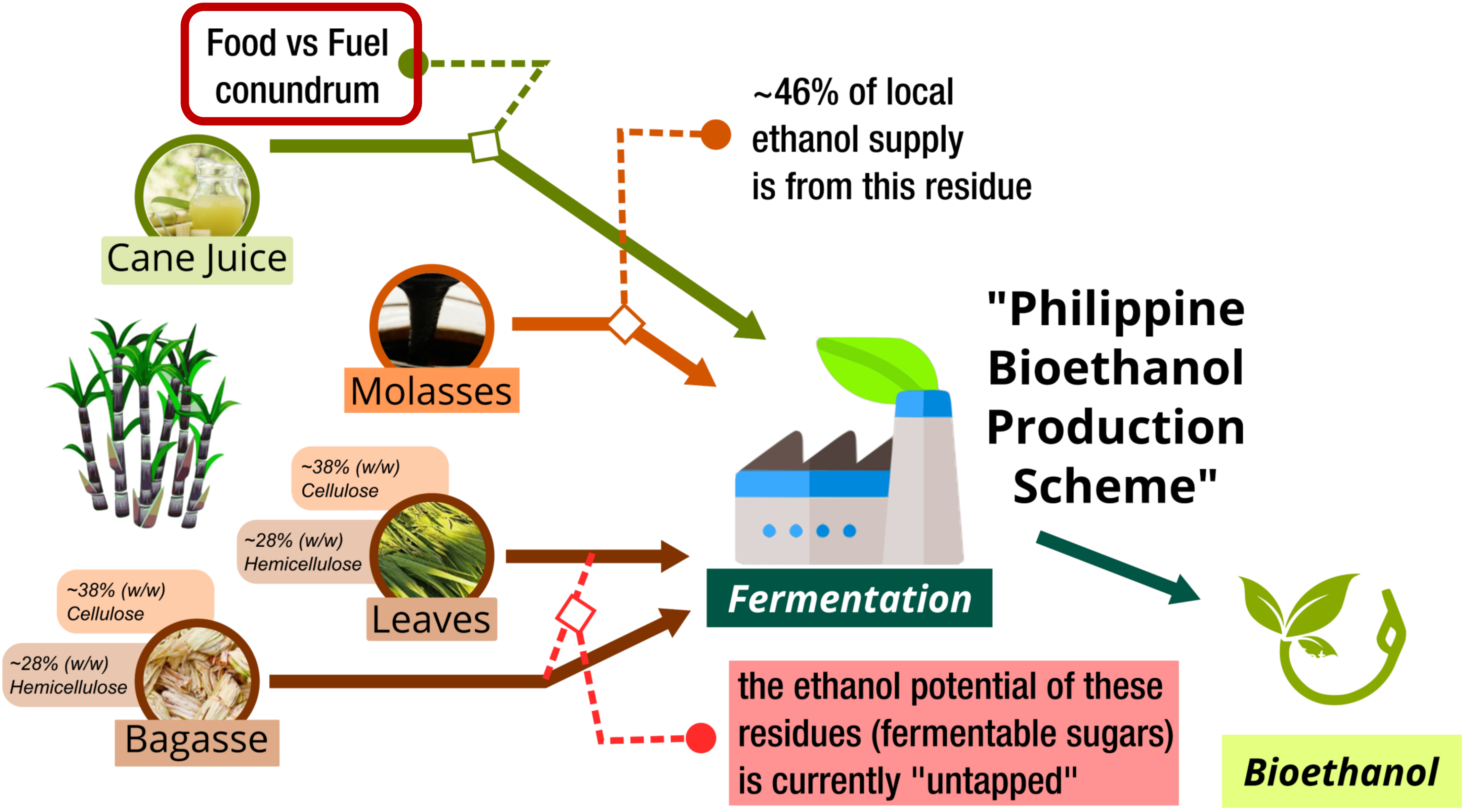
The Biorefinery Concept

"A sustainable processing of biomass into a spectrum of marketable products (food, feed, materials, chemicals) and energy (fuels, power, heat)."

-IEA Bioenergy Task 42
On " Biorefinery Definition"

The Biorefinery Concept





The Biorefinery: Looking at the Philippine Bioethanol Scenario

The Problem

10 %v/v blending of bioethanol

by 2020 →

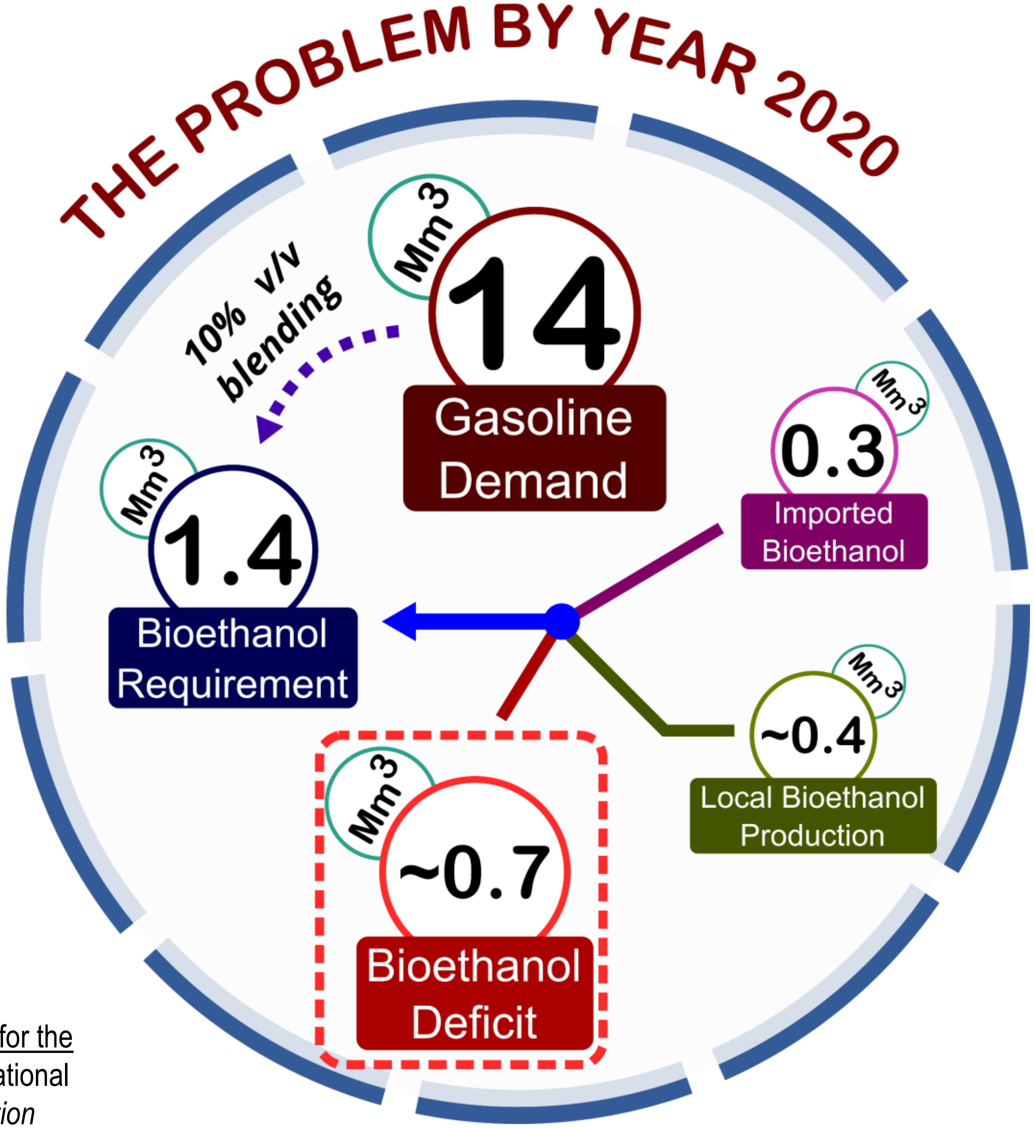
20 %v/v blending of bioethanol

RA 9367

BIOFUELS ACT OF 2006

NATIONAL BIOFUELS BOARD (NBB)

Department of Energy



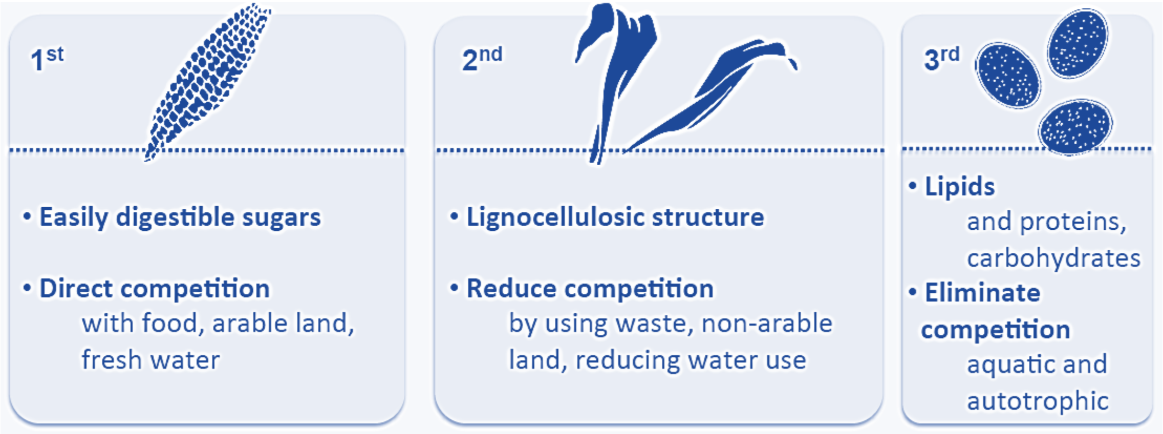
Orilla, J. W. A., A. W. Go, A. T. Conag, A. S. Toledo, Y. H. Ju and I. D. F. Tabañag (2019). Valorization of Sugarcane Residues for the Production of Bioethanol in the Philippine Context: A Retrospective Assessment from 2007-2017 and Future Challenges. 2019 National Research Council of the Philippines (NRCP) Annual Scientific Conference, Manila, Philippines. *Manuscript Submitted for Publication*

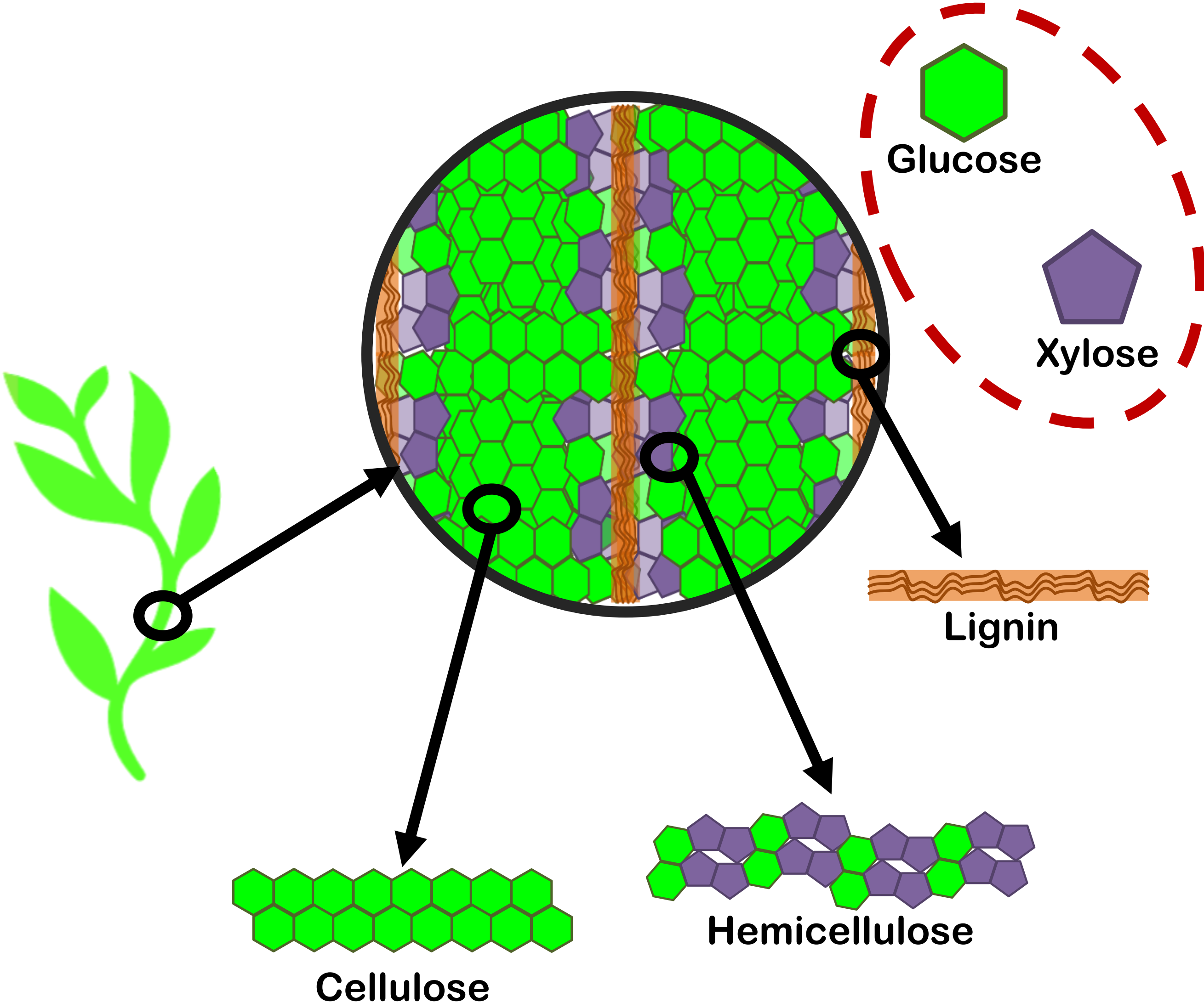
Outline of the Presentation

Biomass Feedstocks

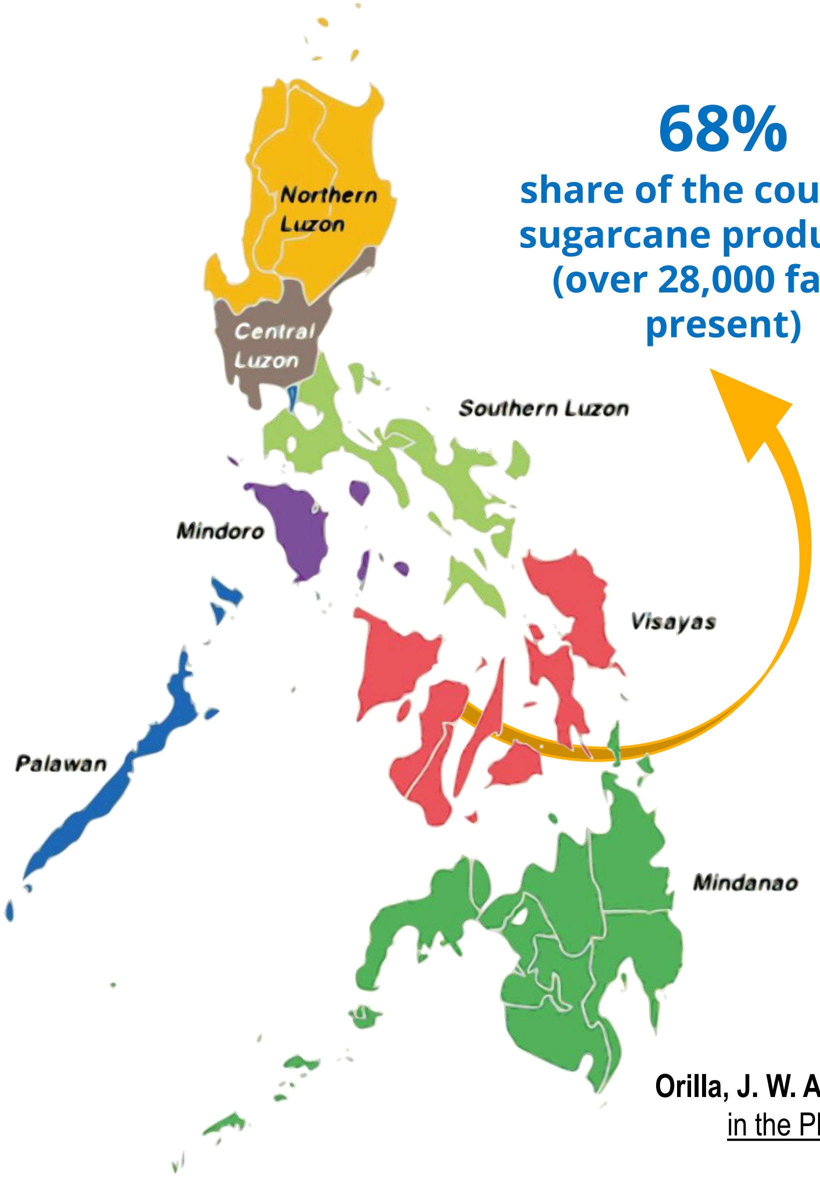
*The **Starting Point** in unlocking our potential to establish a **Bio-Based Economy***

Biomass Feedstocks and their Classification



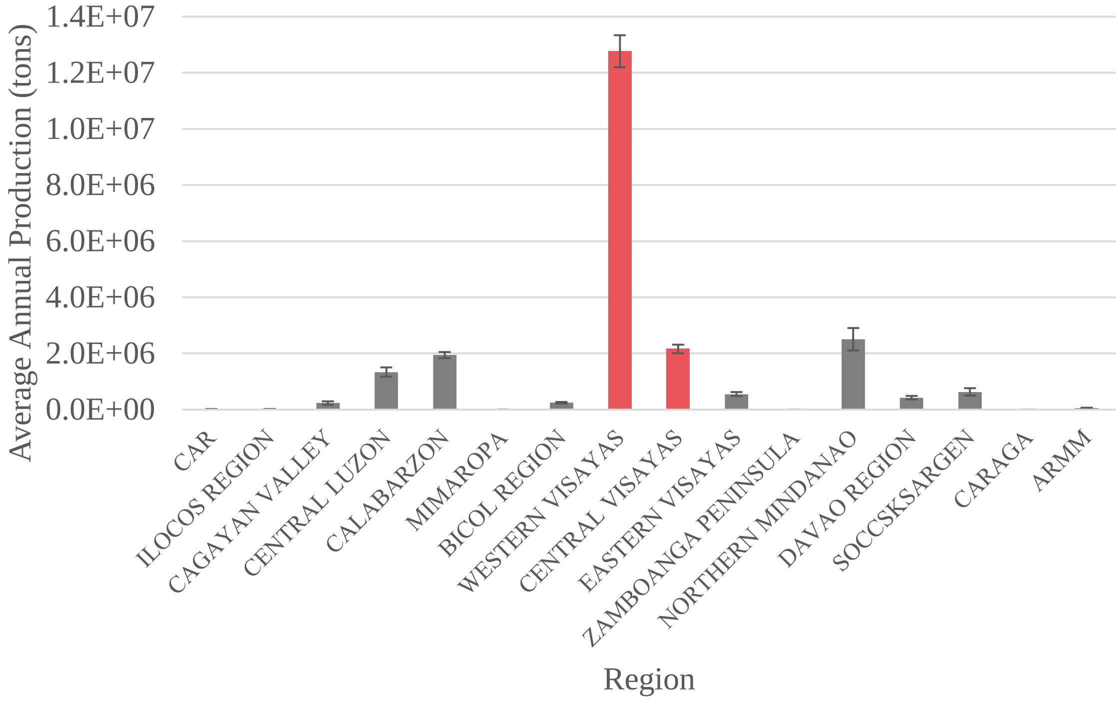


Looking at what we have…



68%

share of the country's sugarcane production (over 28,000 farms present)



1.06 ± 0.28 Mm³ of ethanol

from SCB recoverable from the Visayas from ~2.16Mt of dry SCB/yr

1.00 ± 0.28 Mm³ of ethanol

from SCL recoverable from the Visayas from ~2.33Mt of dry SCL/yr

Orilla, J. W. A., A. W. Go, A. T. Conag, A. S. Toledo, Y. H. Ju and I. D. F. Tabañag (2019). Valorization of Sugarcane Residues for the Production of Bioethanol in the Philippine Context: A Retrospective Assessment from 2007-2017 and Future Challenges. 2019 National Research Council of the Philippines (NRCP) Annual Scientific Conference, Manila, Philippines. *Manuscript Submitted for Publication*

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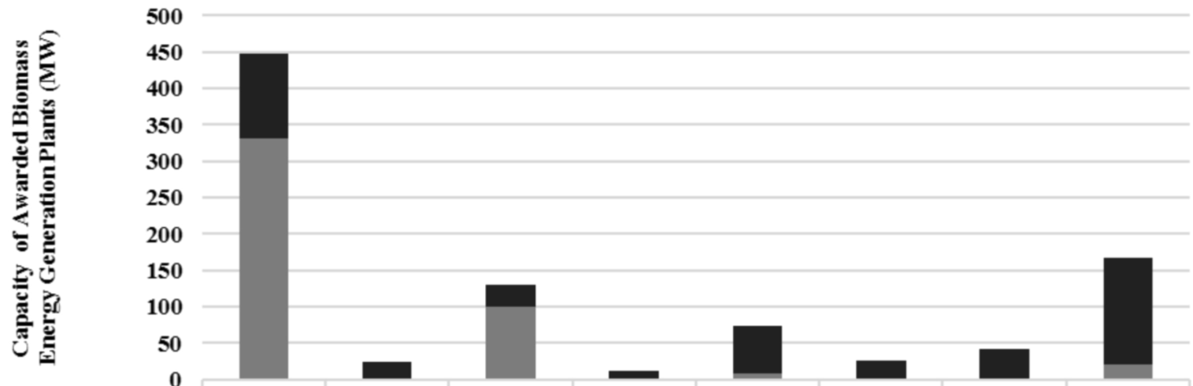


	Sugarcane Bagasse	Sugarcane Trash	Rice husk	Coconut Waste	Multi-Feedstock	Woody Biomass	Napier Grass	Not Specified
Non-Operational	115.8	25	29.75	11.5	65.5	26	41	147.09
Operational	331.28	0	99.7	0	8	0	0	20.024

Feedstock

Figure 4. Capacity and distribution of awarded biomass energy generation plants in the Philippines classified according to the feedstock/fuel used as of 2017 (Data from publushed reports by the Department of Energy of the Philippines (Department of Energy, 2018a, 2018b) were used in generating the figure)

3.43 ± 1.1 Mt of dry SCL/year

3.17 ± 1.1 Mt of dry SCB/year

0.92 ± 0.05 Mt of molasses/year

Percentage of the available SCB used as solid fuel rather than feedstock for bioethanol production **77%**

The Challenge:

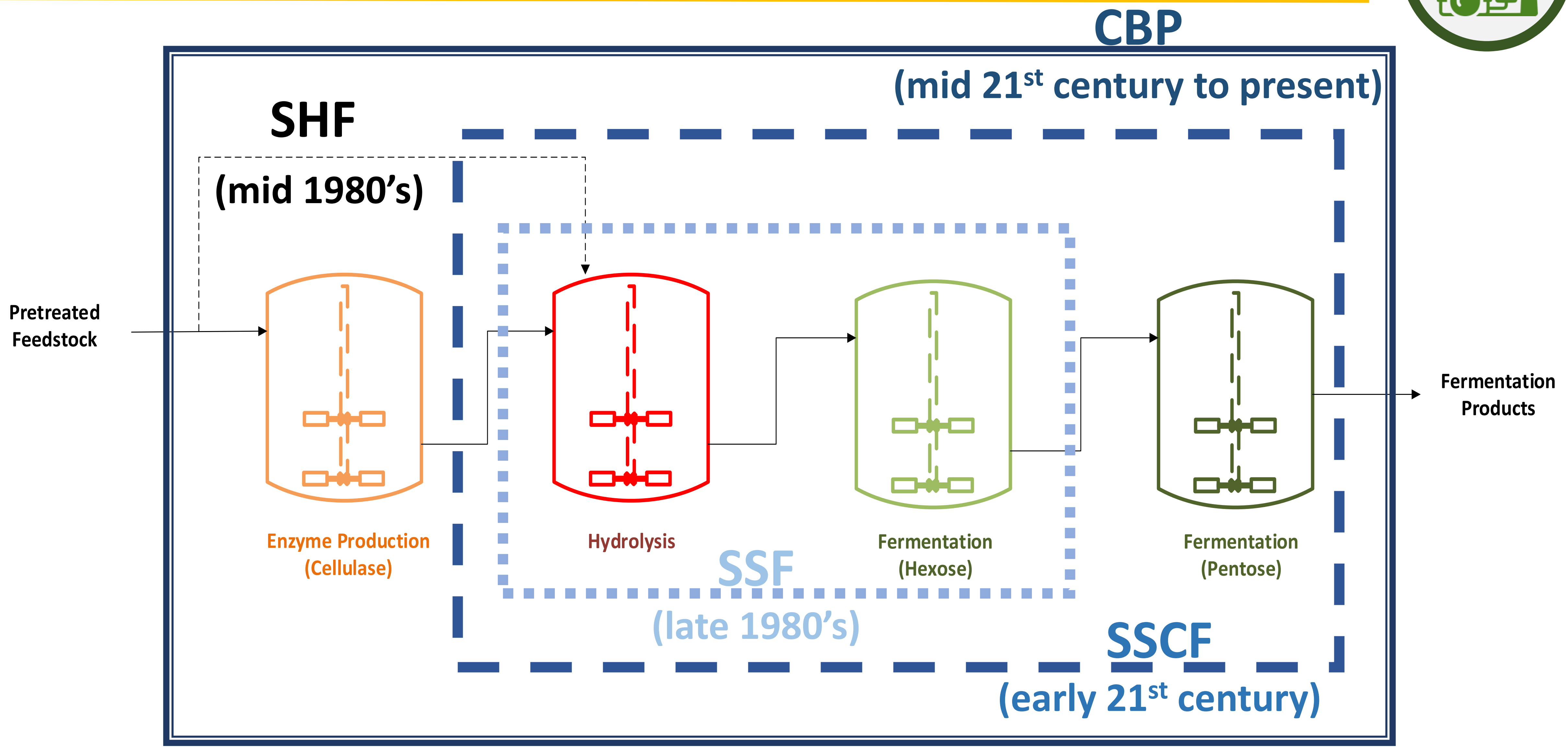
On spreading the awareness that this "underutilized feedstock" can be "valorized" to produce "high-value products" that can set the foundations of a bio-based economy

What are we doing with these sugarcane residues?

Bioprocessing

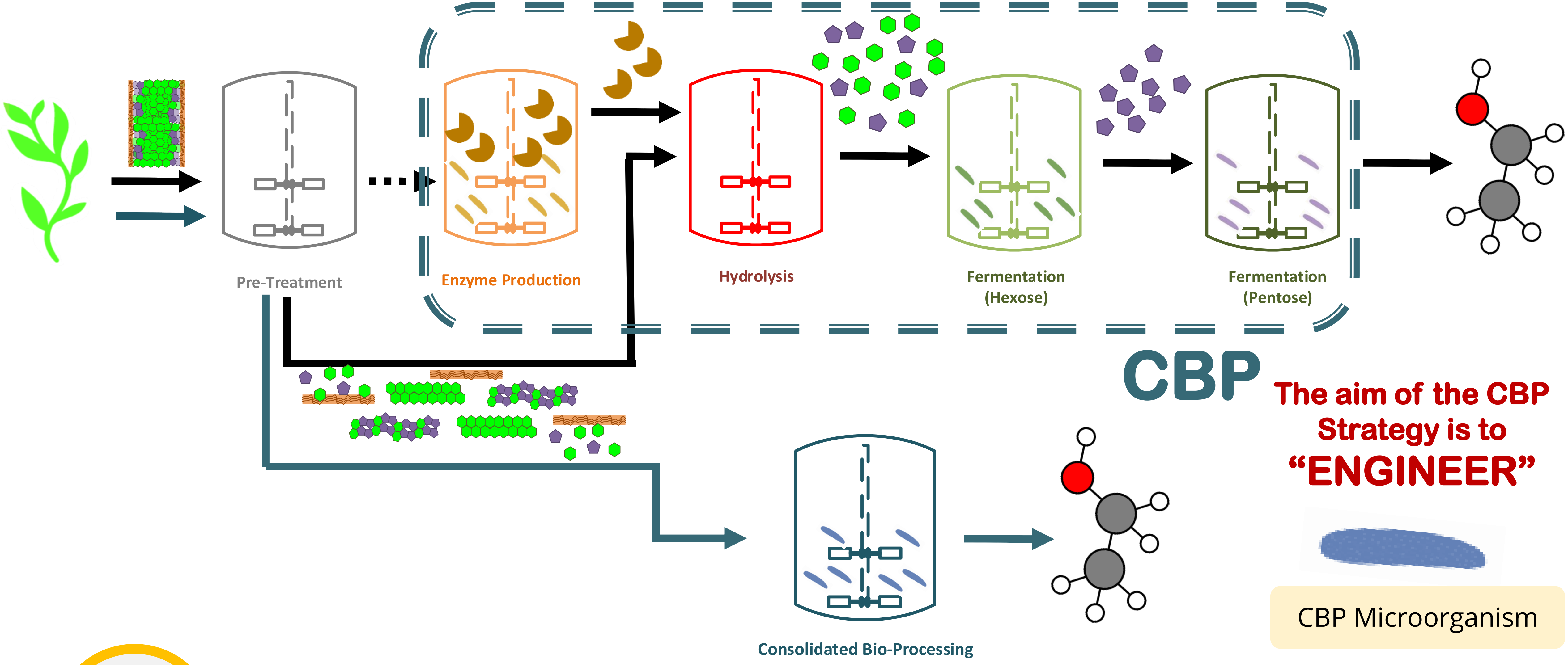
Converting biomass into **platform chemicals** for further production of bio-based products

The Consolidated Bio-Processing Strategy: *A Perspective*



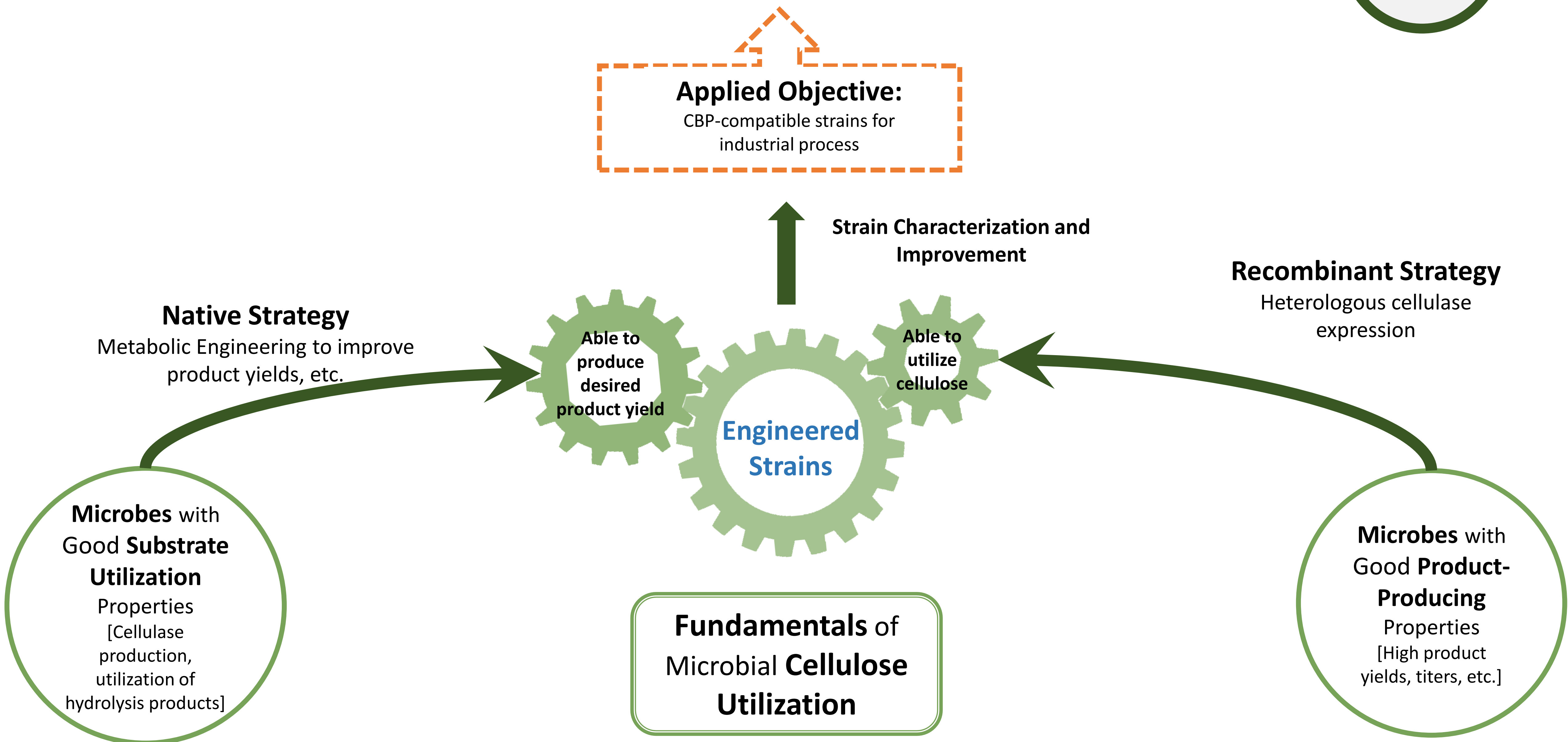
Lynd, L. R., P. J. Weimer, W. H. van Zyl and I. S. Pretorius (2002). "Microbial Cellulose Utilization: Fundamentals and Biotechnology." Microbiology and Molecular Biology Reviews **66**(3): 506-577.

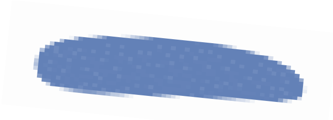
Lynd, L. R., P. J. Weimer, W. H. van Zyl and I. S. Pretorius (2002). "Microbial Cellulose Utilization: Fundamentals and Biotechnology." Microbiology and Molecular Biology Reviews **66**(3): 506-577.



On CBP and it's GOAL

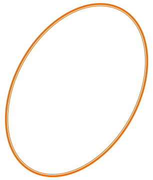
The Qualities of the CBP Microorganism



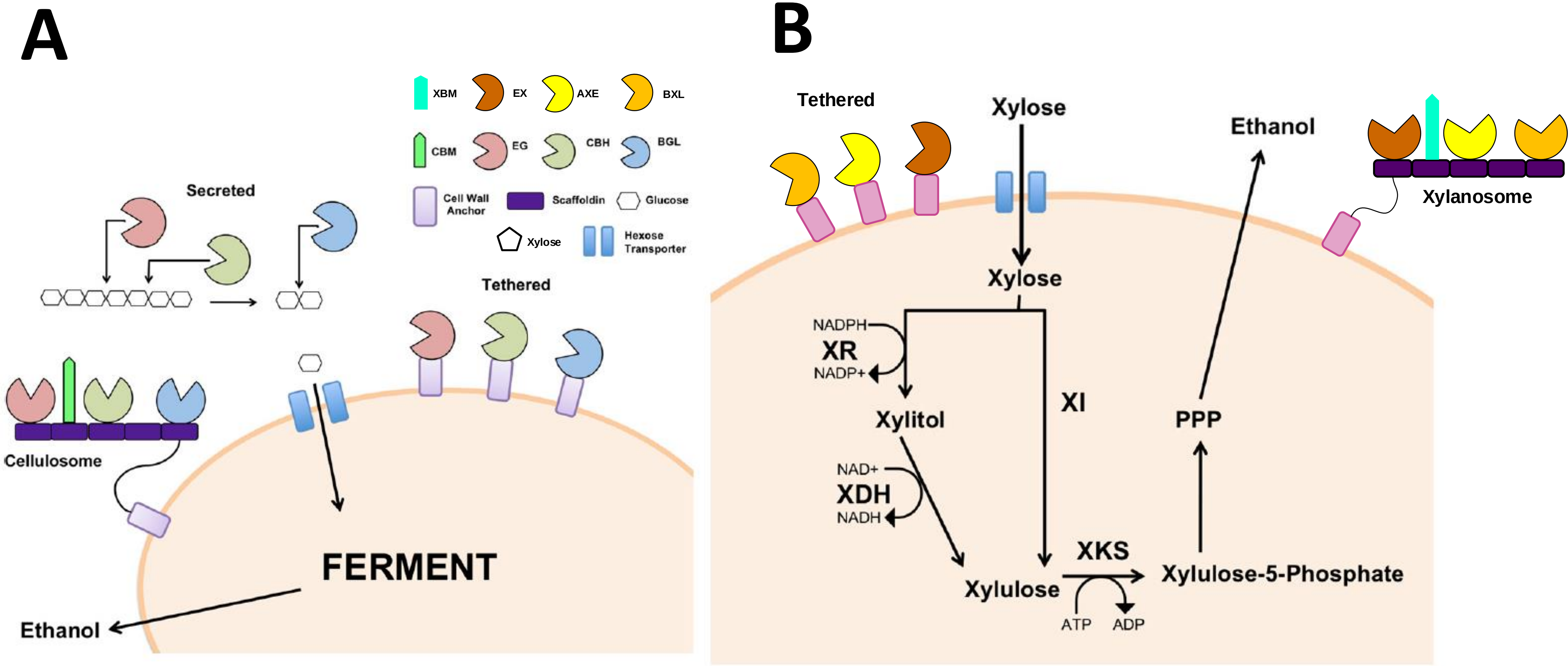


CBP Microorganism

Saccharomyces Cerevisiae



YSc



Engineering Yeast for Cellulose **(A)** and Hemicellulose **(B)** Utilization

Kricka, W., J. Fitzpatrick and U. Bond (2014). "Metabolic engineering of yeasts by heterologous enzyme production for degradation of cellulose and hemicellulose from biomass: a perspective." Front Microbiol **5**: 174.

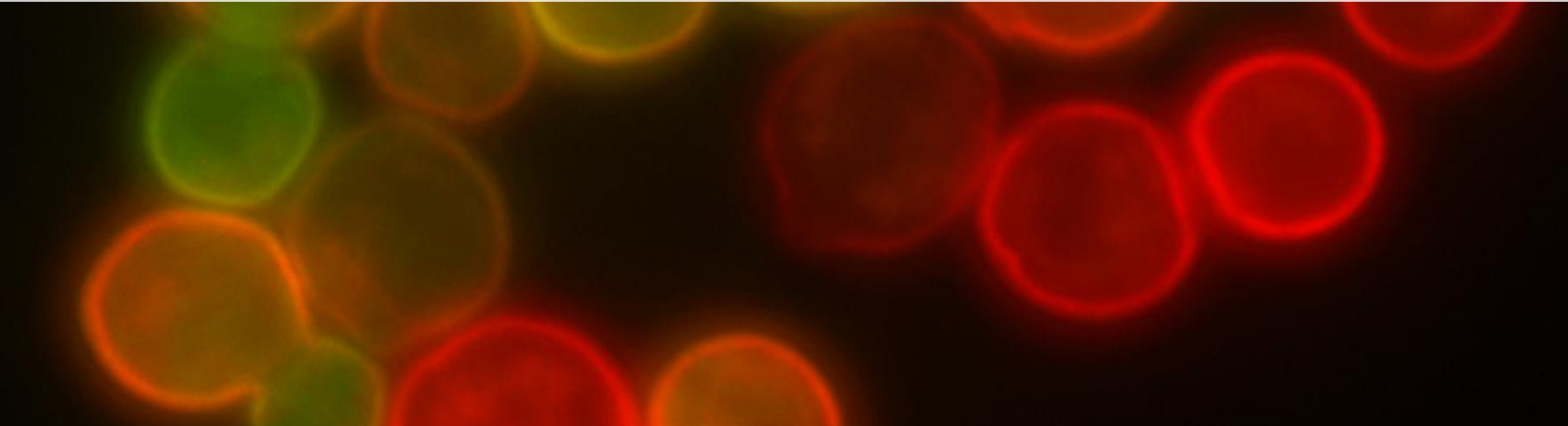
Lignocellulosic conversion via the CBP Microorganism

From lignocellulosic biomass to bioethanol …

Yeast Cell Surface Display in Lignocellulosic Bioethanol Production

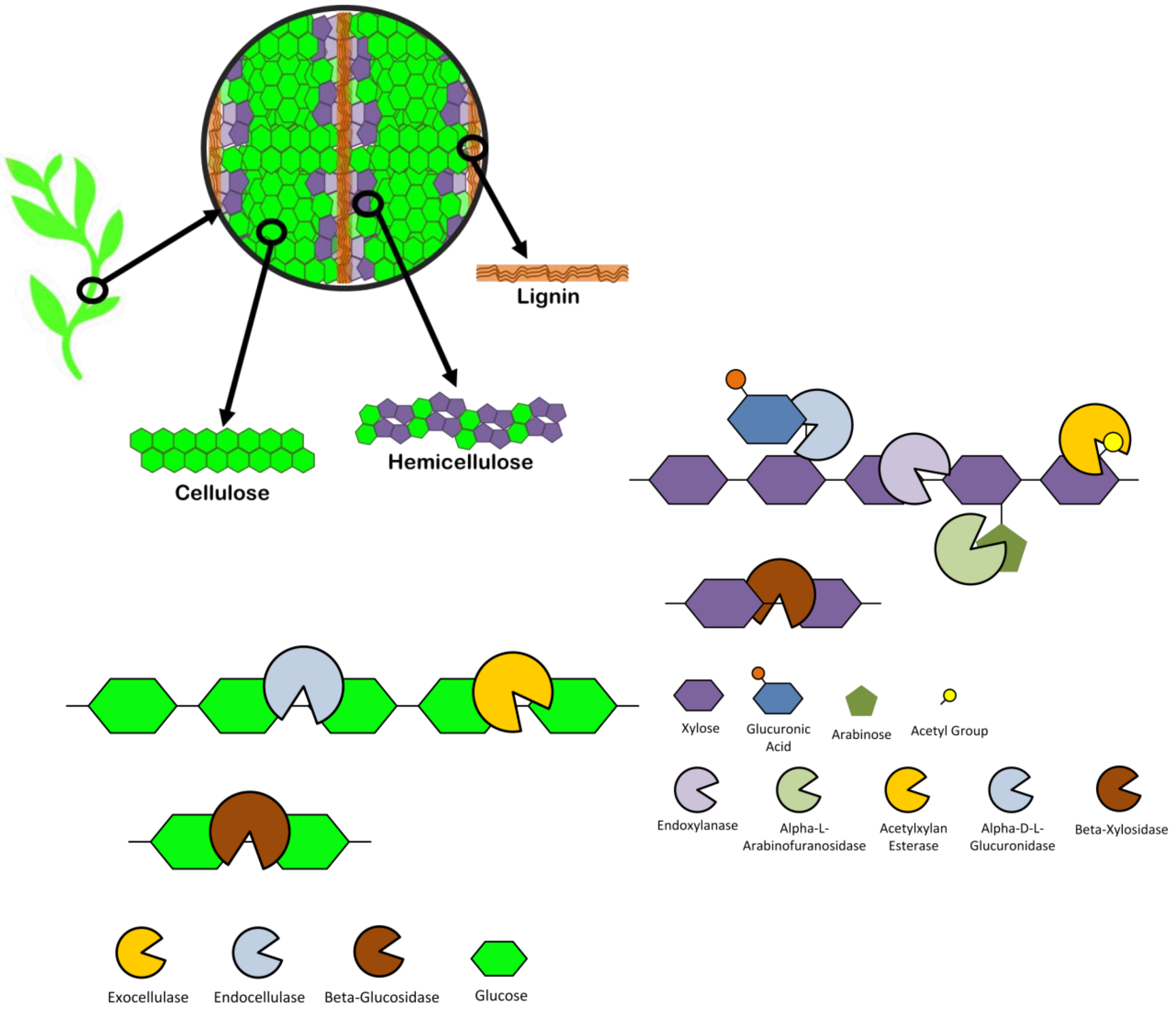
"this field has elevated the status of S. cerevisiae as a novel and attractive microorganism to be a platform for enzyme immobilization which enables it to target nonconventional substrates and at the same time, renewable self-immobilized biocatalysts (a.k.a whole-cell biocatalysts) ."

-Ueda and Tanaka 2000
J. Biosci. Bioeng

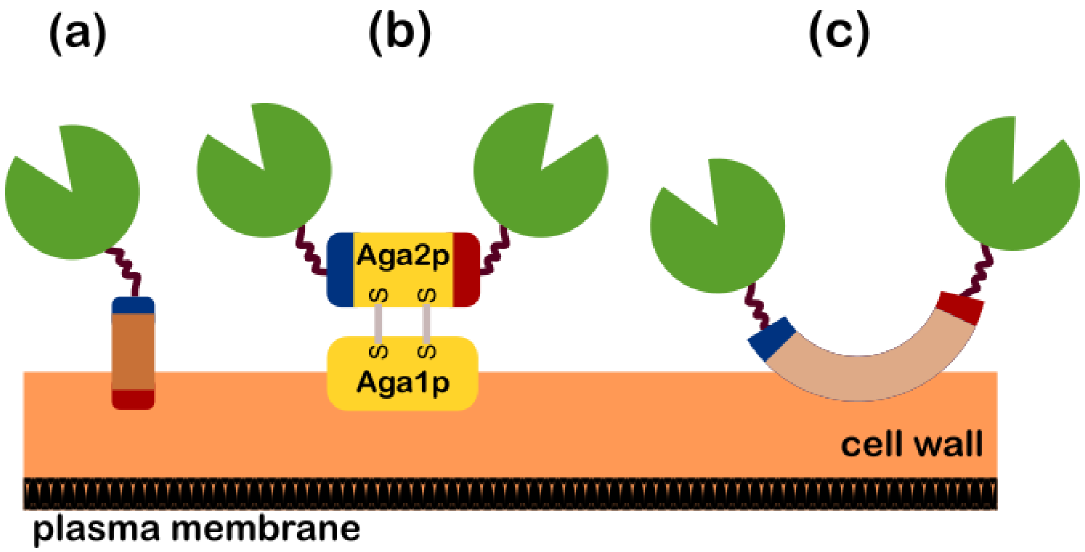


Why Yeast Cell Surface Display???

Tabañag, I. D. F. and S. L. Tsai (2017). Biological Strategies in Nanobiocatalyst Assembly. Biocatalysis and Nanotechnology. P. Grunwald, CRC Press: 151-241.



The Inherent Surface Display Systems in S. cerevisiae



Common anchor protein systems used in yeast surface display: (**a**) N-terminal fused anchor proteins (SAG1, SED1, or CWP2); (**b**) the a-agglutinin display system; and, (**c**) the Flo1p-based display system. Anchor proteins: N-Terminal depicted in blue, and C-terminal depicted in red.
(https://www.yeastgenome.org/)

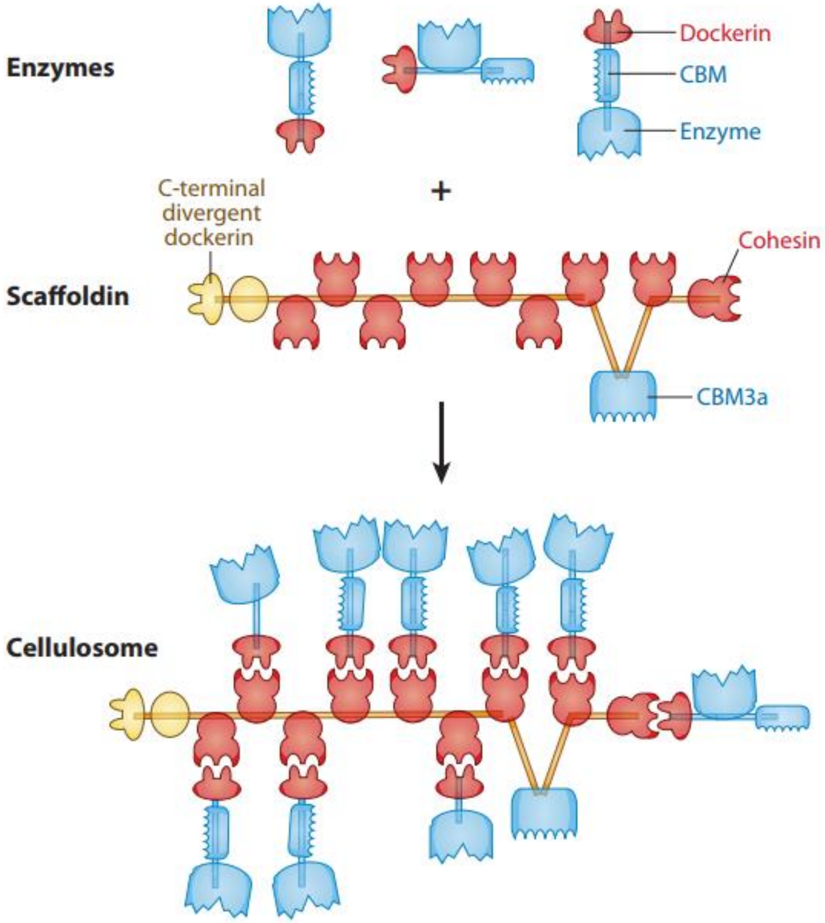
Kricka, W., J. Fitzpatrick and U. Bond (2014). "Metabolic engineering of yeasts by heterologous enzyme production for degradation of cellulose and hemicellulose from biomass: a perspective." Front Microbiol **5**: 174.

On Yeast Surface Display and Lignocellulose Conversion - I

On the discovery of the structured assembly of cellulolytic enzymes on *Clostridium thermocellum*

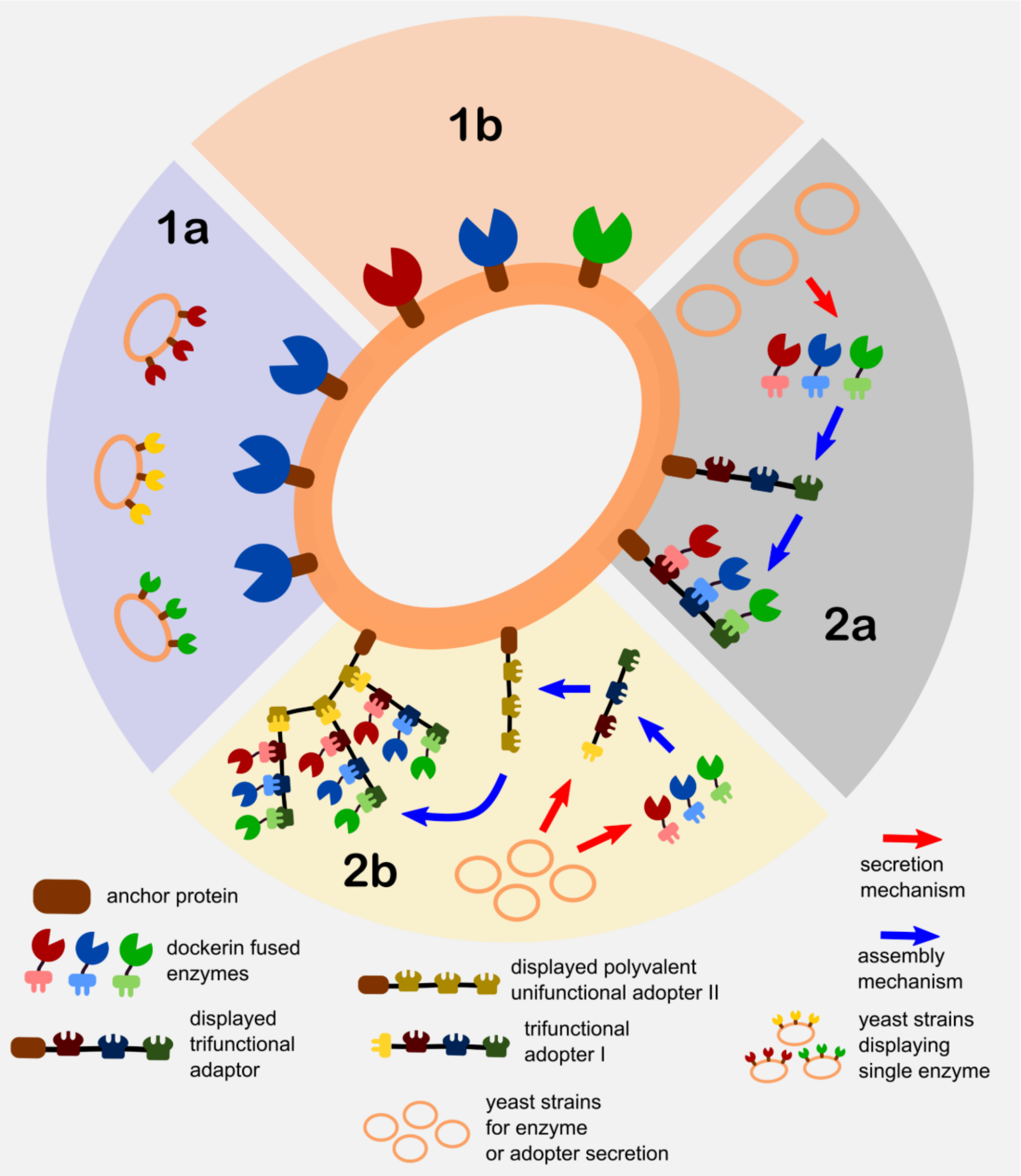
At the height of **biomimicry**, this discovery has been exploited for its functional assembly characteristic

THE CELLULOSOME STRUCTURE



Fontes, C. M. and H. J. Gilbert (2010). "Cellulosomes: highly efficient nanomachines designed to deconstruct plant cell wall complex carbohydrates." Annu Rev Biochem **79**(1): 655-681.

How is this platform exploited for Bioethanol Production?



Direct Yeast Surface Display (DYSD)

directly involves the fusion of the enzyme of interest/s to the yeast anchor protein for it to be expressed and localize on the cell surface

1a – Single Enzyme Display Consortium (SEDC)

1b – Co-Display (CD)

Juxtaposed Assembly of Non-Native Adaptors and Secreted Enzymes on Yeast (JANNASEY)

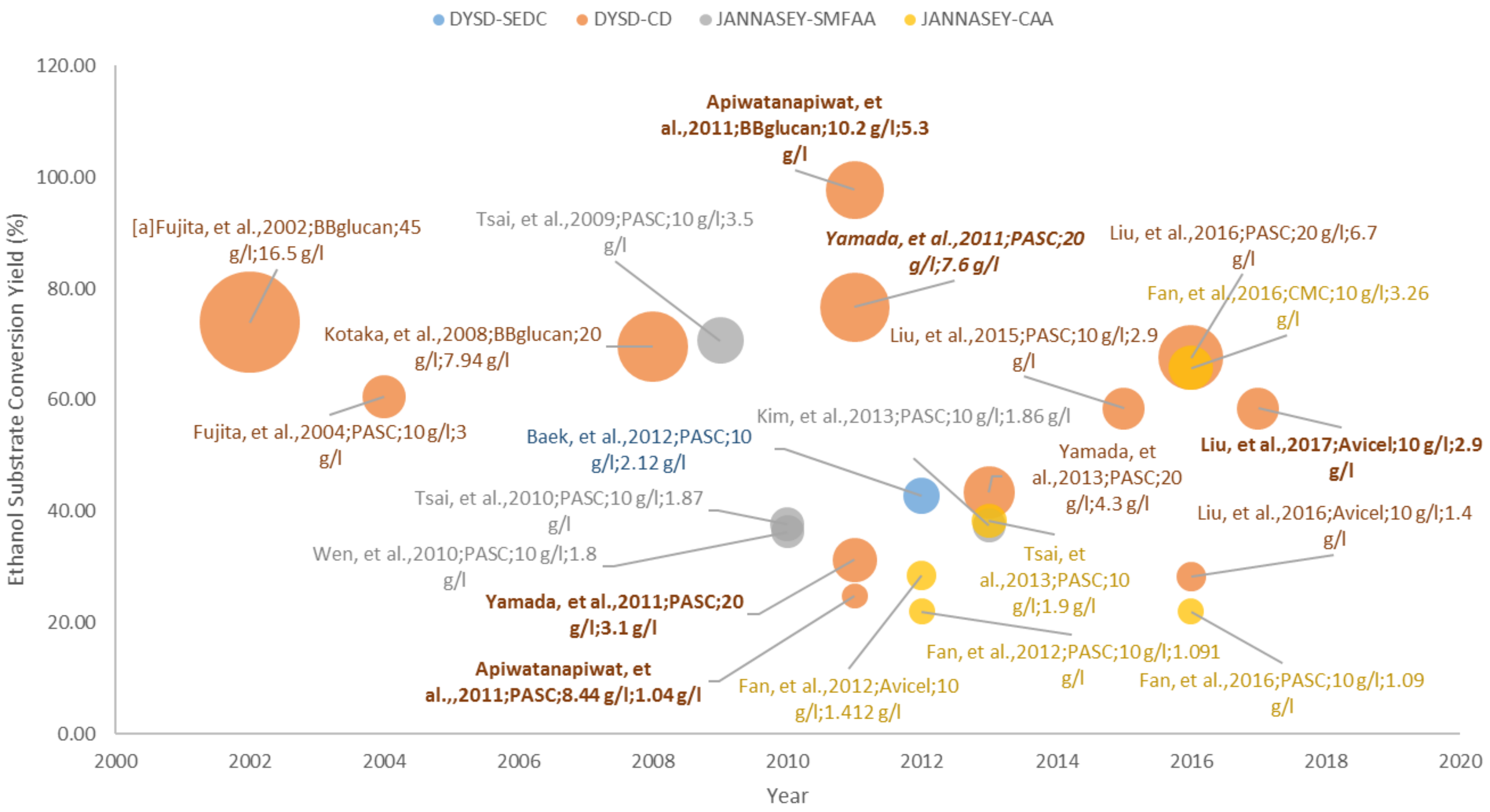
developed based on the biomimicry of the "cellulosome" machinery of several anaerobic cellulose-consuming microorganisms (e.g., clostridia, ruminal bacteria)

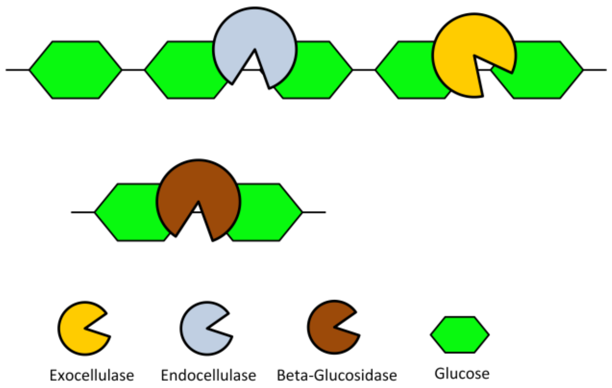
2a – Single Multi-Functional Adaptor Assembly (SMFAA)

2b – Complex Adaptor Assembly (CAA)

Tabañag, I. D. F., I.-M. Chu, Y.-H. Wei and S.-L. Tsai (2018). "The Role of Yeast-Surface-Display Techniques in Creating Biocatalysts for Consolidated BioProcessing." Catalysts **8**(3): 94.

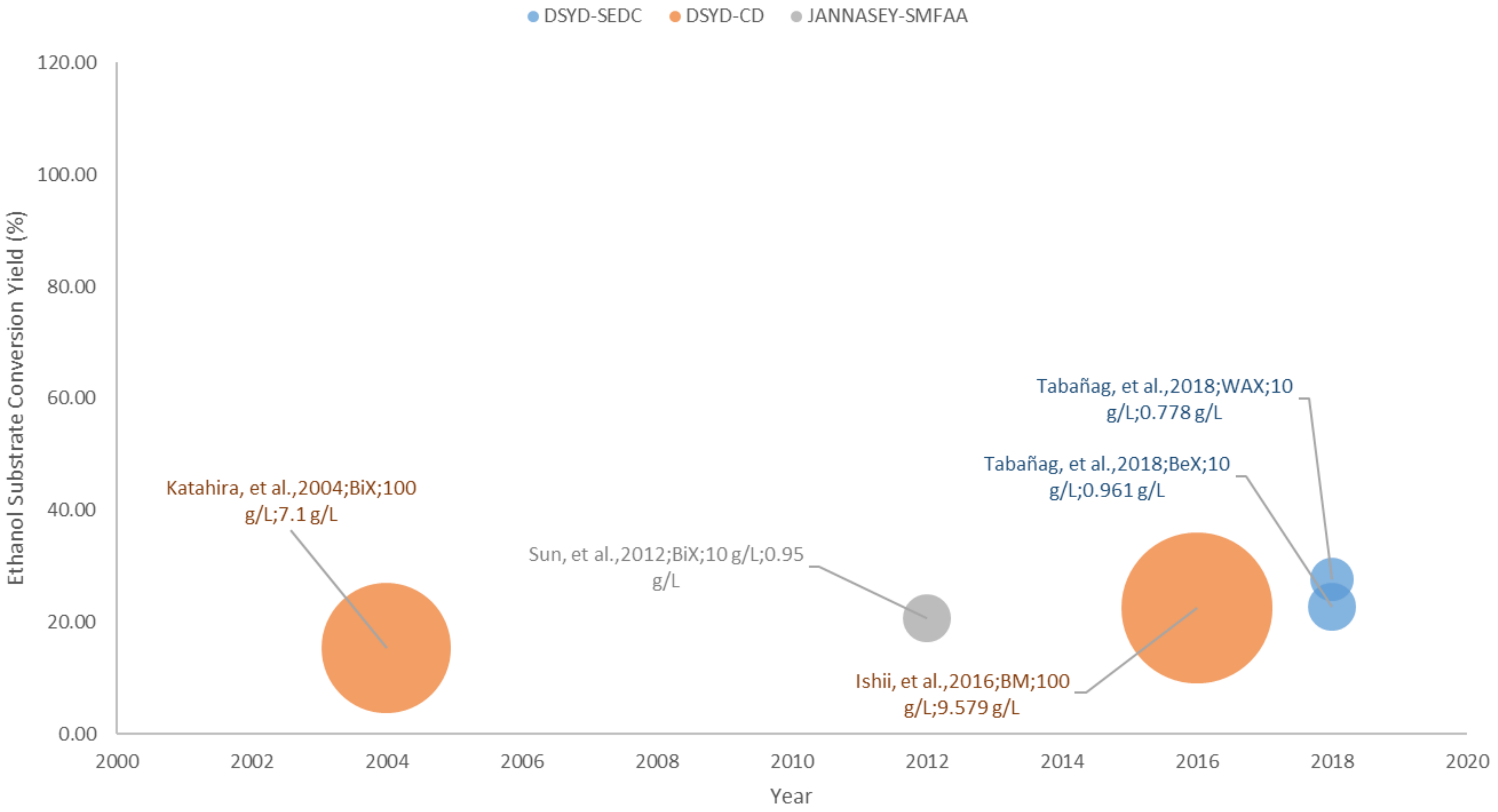
Visualization of the status and development of yeast surface display for Cellulosic Bioethanol Production. Bubble sizes represent ethanol titer. Data labels are formatted as follows: (Reference (author-date); Substrate; Substrate Concentration; Ethanol Titer). For the DSYD-CD data set: labels in boldface represent cellulase ratio control via delta-integration; labels in italic represent utilization of diploid host strains.



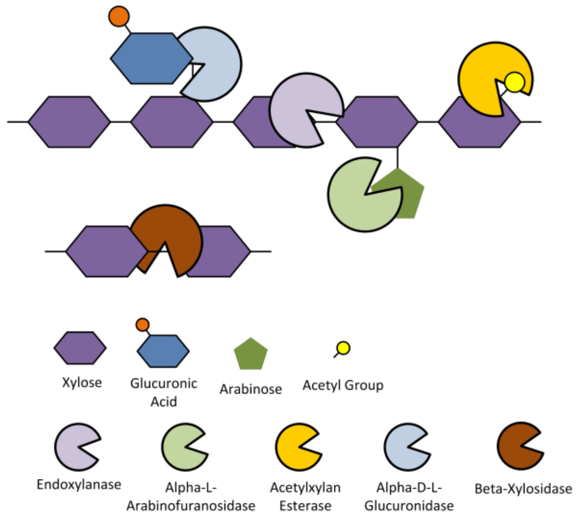


Tabañag, I. D. F., I.-M. Chu, Y.-H. Wei and S.-L. Tsai (2018). "The Role of Yeast-Surface-Display Techniques in Creating Biocatalysts for Consolidated BioProcessing." Catalysts **8**(3): 94.

YSD and Bioethanol Production from Pure Polysaccharide Substrates



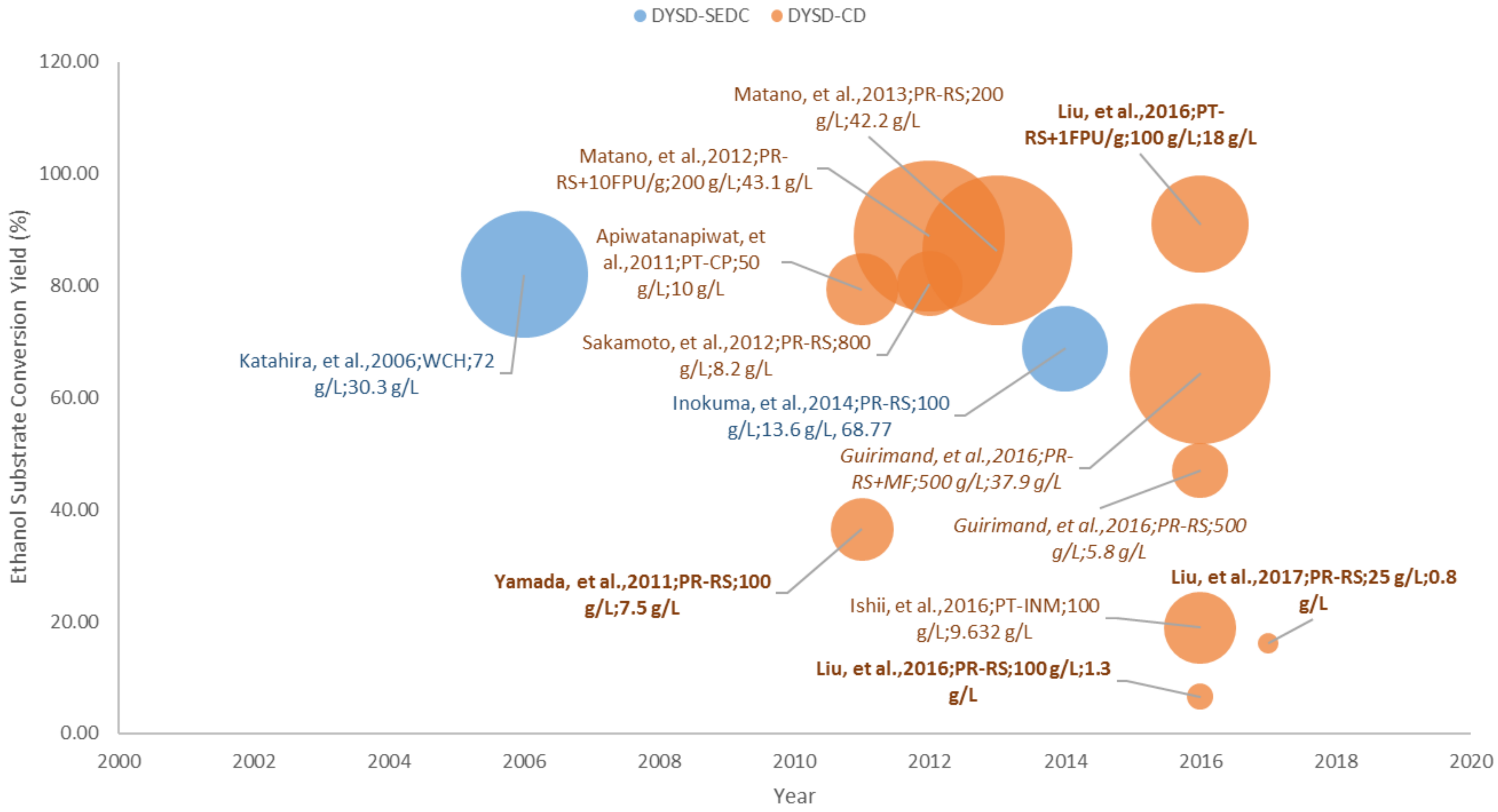
Visualization of the status and development of yeast surface display for Hemicellulosic Bioethanol Production. Bubble sizes represent ethanol titer. Data labels are formatted as follows: (Reference (author-date); Substrate; Substrate Concentration; Ethanol Titer).



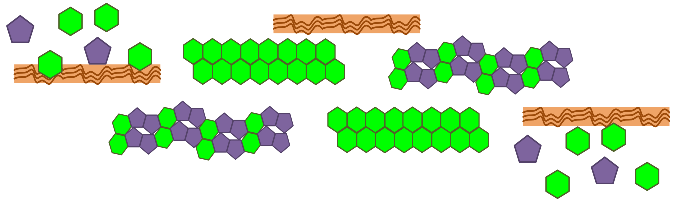
Tabañag, I. D. F., I.-M. Chu, Y.-H. Wei and S.-L. Tsai (2018). "The Role of Yeast-Surface-Display Techniques in Creating Biocatalysts for Consolidated BioProcessing." Catalysts **8**(3): 94.

YSD and Bioethanol Production from Pure Polysaccharide Substrates

YSD and Bioethanol Production from Pre-Treated LB Substrates

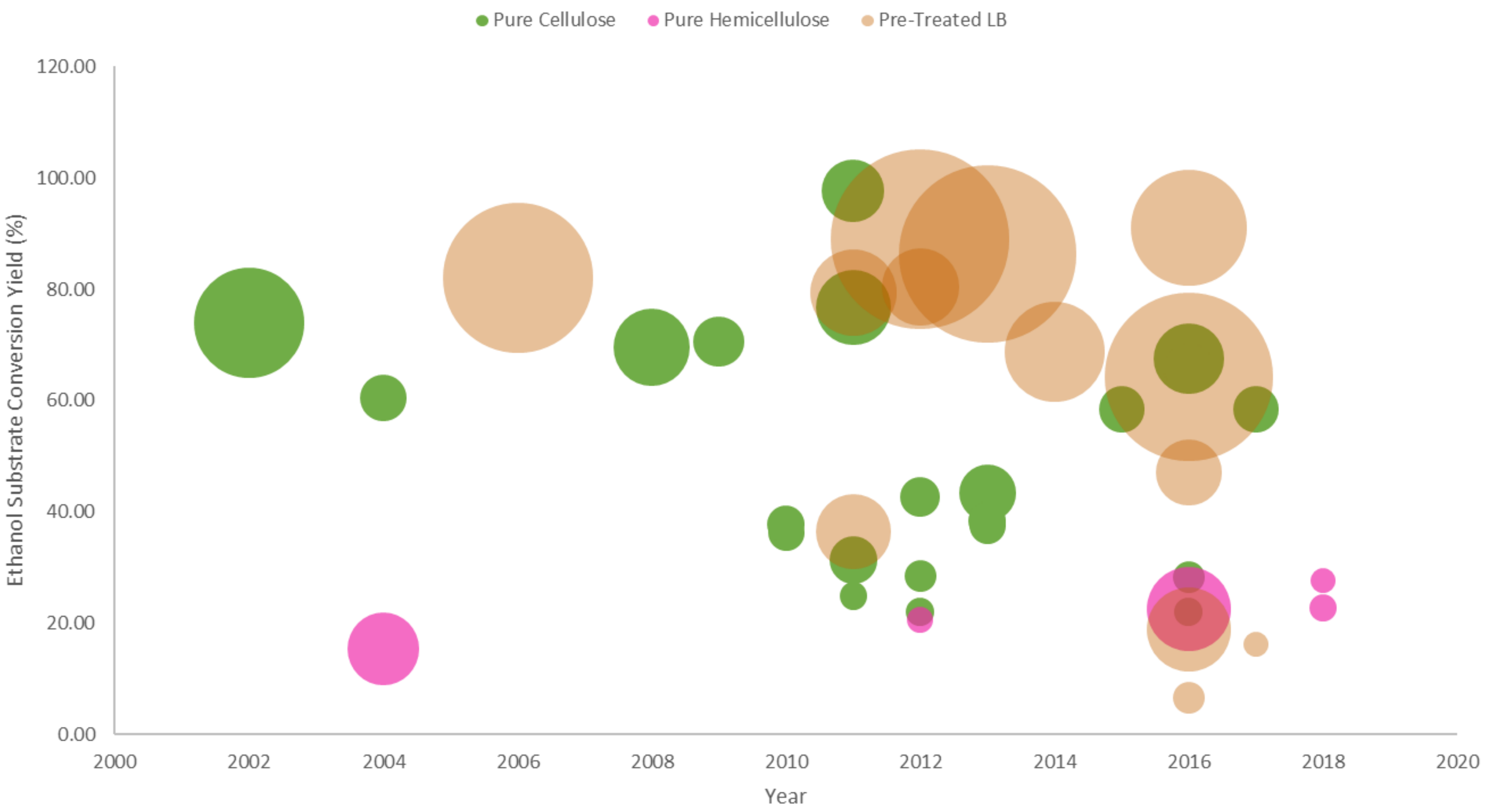


Visualization of the status and development of yeast surface display for Bioethanol Production from Pre-Treated Lignocellulosic Biomass. Bubble sizes represent ethanol titer. Data labels are formatted as follows: (Reference (author-date); Substrate; Substrate Concentration; Ethanol Titer). For DSYD-CD data sets: labels in boldface represent cellulase ratio control via delta-integration; labels in italic represent the xylitol fermentation parameters instead of ethanol.

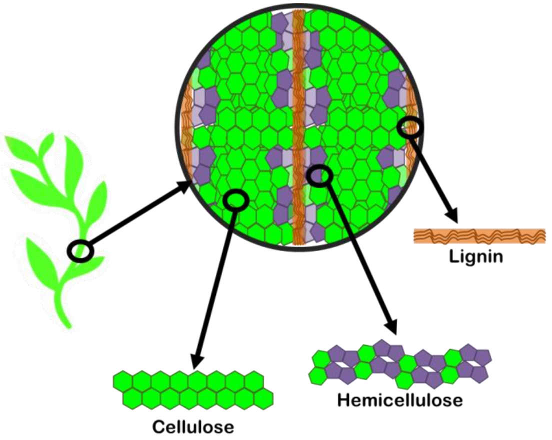


Tabañag, I. D. F., I.-M. Chu, Y.-H. Wei and S.-L. Tsai (2018). "The Role of Yeast-Surface-Display Techniques in Creating Biocatalysts for Consolidated BioProcessing." Catalysts **8**(3): 94.

A perspective on the extent of YSD application to various substrates



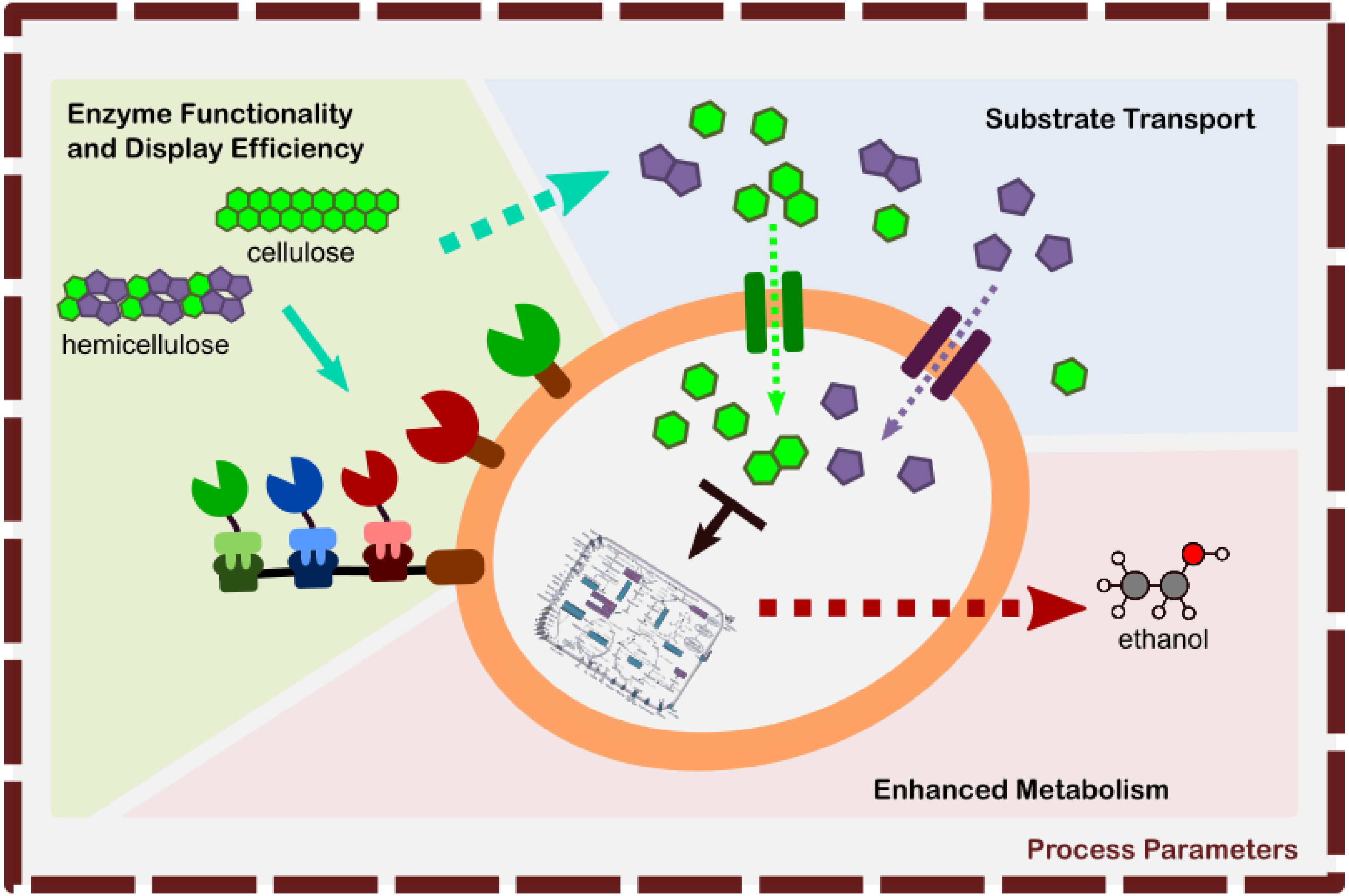
Visualization of the status and development of yeast surface display for Bioethanol Production from Pre-Treated Lignocellulosic Biomass. Bubble sizes represent ethanol titer. Data labels are formatted as follows: (Reference (author-date); Substrate; Substrate Concentration; Ethanol Titer). For DSYD-CD data sets: labels in boldface represent cellulase ratio control via delta-integration; labels in italic represent the xylitol fermentation parameters instead of ethanol.



Tabañag, I. D. F., I.-M. Chu, Y.-H. Wei and S.-L. Tsai (2018). "The Role of Yeast-Surface-Display Techniques in Creating Biocatalysts for Consolidated BioProcessing." Catalysts **8**(3): 94.

“The Role of Yeast Surface Display in Bioethanol Production from Lignocellulosic Substrates”

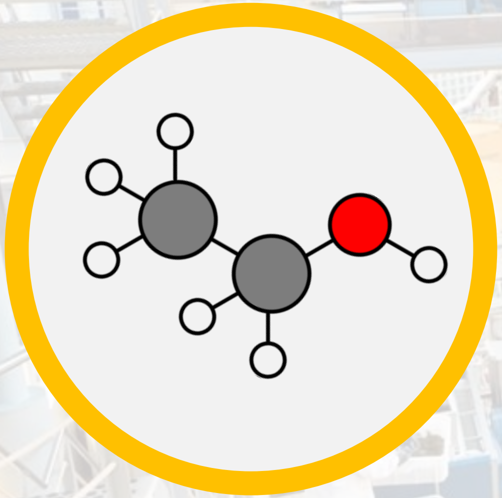
On the Variables that Affect the Strain Performance



Tabañag, I. D. F., I.-M. Chu, Y.-H. Wei and S.-L. Tsai (2018). "The Role of Yeast-Surface-Display Techniques in Creating Biocatalysts for Consolidated BioProcessing." <u>Catalysts</u> **8**(3): 94.

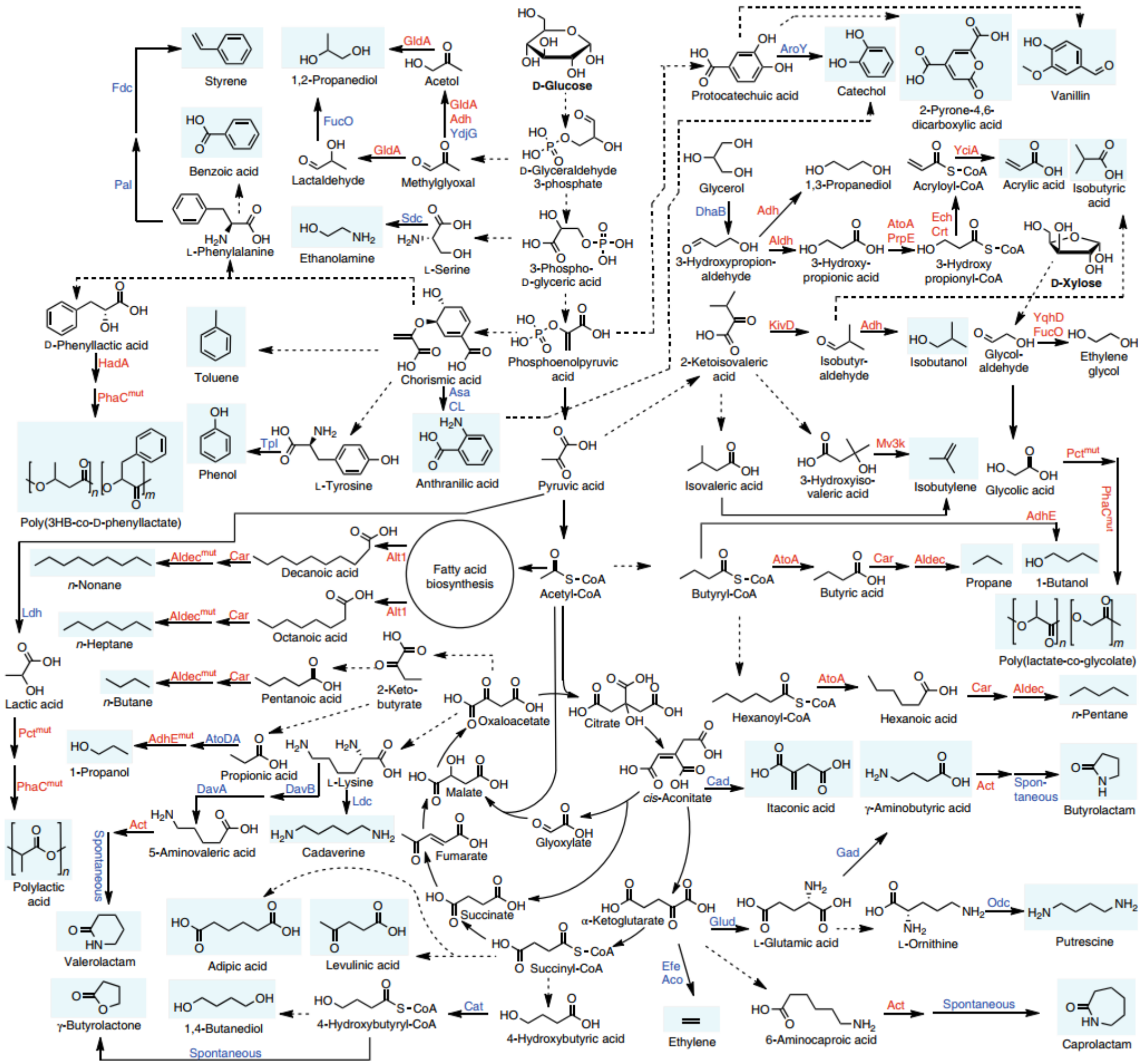
On the avenues of improvement

Bio-Based Platform Chemicals



A prelude to **"Bio-based Products"**

Taking a peek at the bio-based platform chemicals production



Industrial chemicals and materials produced using only biological reactions. This simplified map covers 92 out of 435 chemicals and materials (boxed) presented in the poster (downloadable through the QR code). Solid and dotted arrows indicate single and multiple reaction steps, respectively. The abbreviated enzyme names indicate enzymes with a single substrate specificity (blue) and those with a broad substrate range (red). Glucose is used as a representative carbon source towards the production of industrially relevant chemicals and materials. Xylose is another carbon source that is also often considered for the production of ethylene glycerol and poly(lactate-co-glycolate). The fatty acid biosynthetic pathway is presented as a circle to indicate a series of fatty acid chain elongation reactions.

Bio-Based Chemicals Map

Bio-Based Chemicals Paper

Lee, S. Y., H. U. Kim, T. U. Chae, J. S. Cho, J. W. Kim, J. H. Shin, D. I. Kim, Y.-S. Ko, W. D. Jang and Y.-S. Jang (2019). "A comprehensive metabolic map for production of bio-based chemicals." Nature Catalysis **2**(1): 18-33.

The Chemical Engineer and his/her role in our transition towards the Bio-based Economy

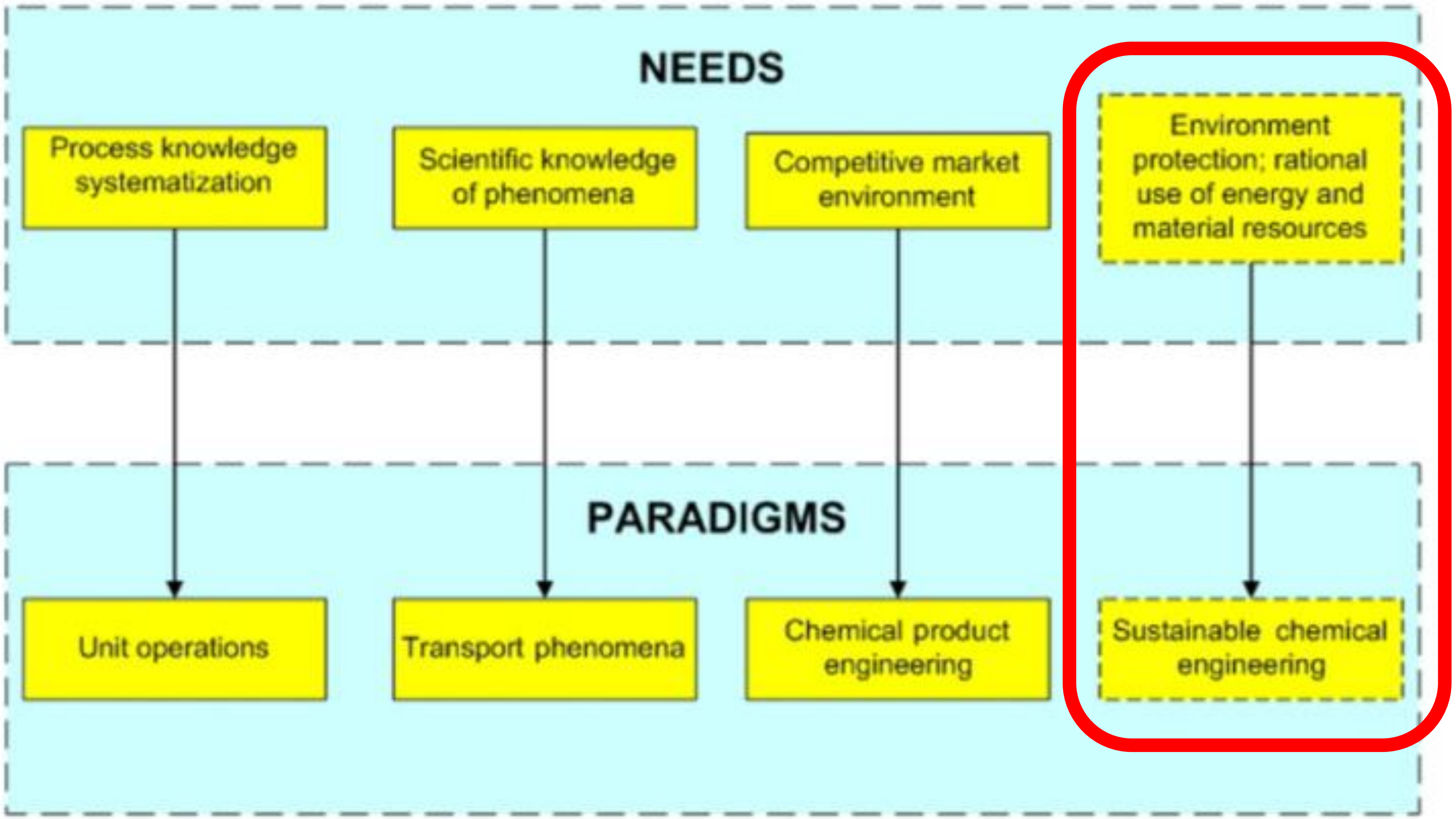
"the mission and essence of chemical engineering is to come up with processes to make materials wanted by man-new or improved processes to replace older less efficient ones, and processes to make completely new materials. In a nutshell we are the chefs of science and technology, and as I put it here, it's a two-step affair-conceiving or dreaming up a scheme, and then making it come real. "

-Octave Levenspiel, 1988

In "Chemical Engineering's Grand Adventure"

The Chemical Engineering Paradigms

The needs in the evolution of chemical engineering and its corresponding paradigms

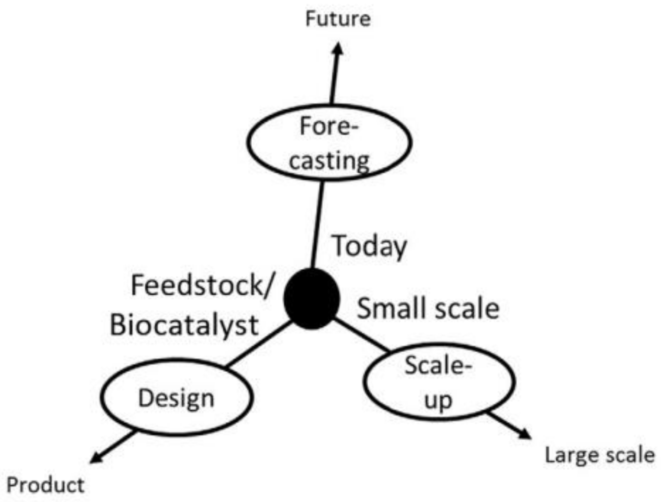


Woinaroschy, A. (2016). "A paradigm-based evolution of chemical engineering." Chinese Journal of Chemical Engineering **24**(5): 553-557.

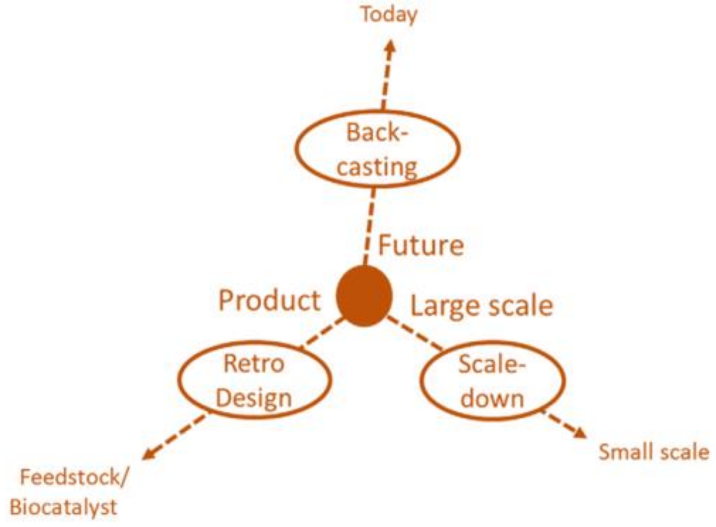
Noorman, H. J. and J. J. Heijnen (2017). "Biochemical engineering's grand adventure." Chemical Engineering Science **170**: 677-693.

"The mission and essence of biochemical engineering is to deliver products that are desired by humanity, from processes where microorganisms, enzymes and/or cell lines convert renewable feedstocks, or intermediates derived from them, into added-value products in a chain of operations. Similar to chemical engineering, also biochemical engineering is a two-step activity: first conceiving a design, and then putting it into reality."

Bioprocess Design in the context of 'transitioning from a fossil-based to a bio-based economy…'

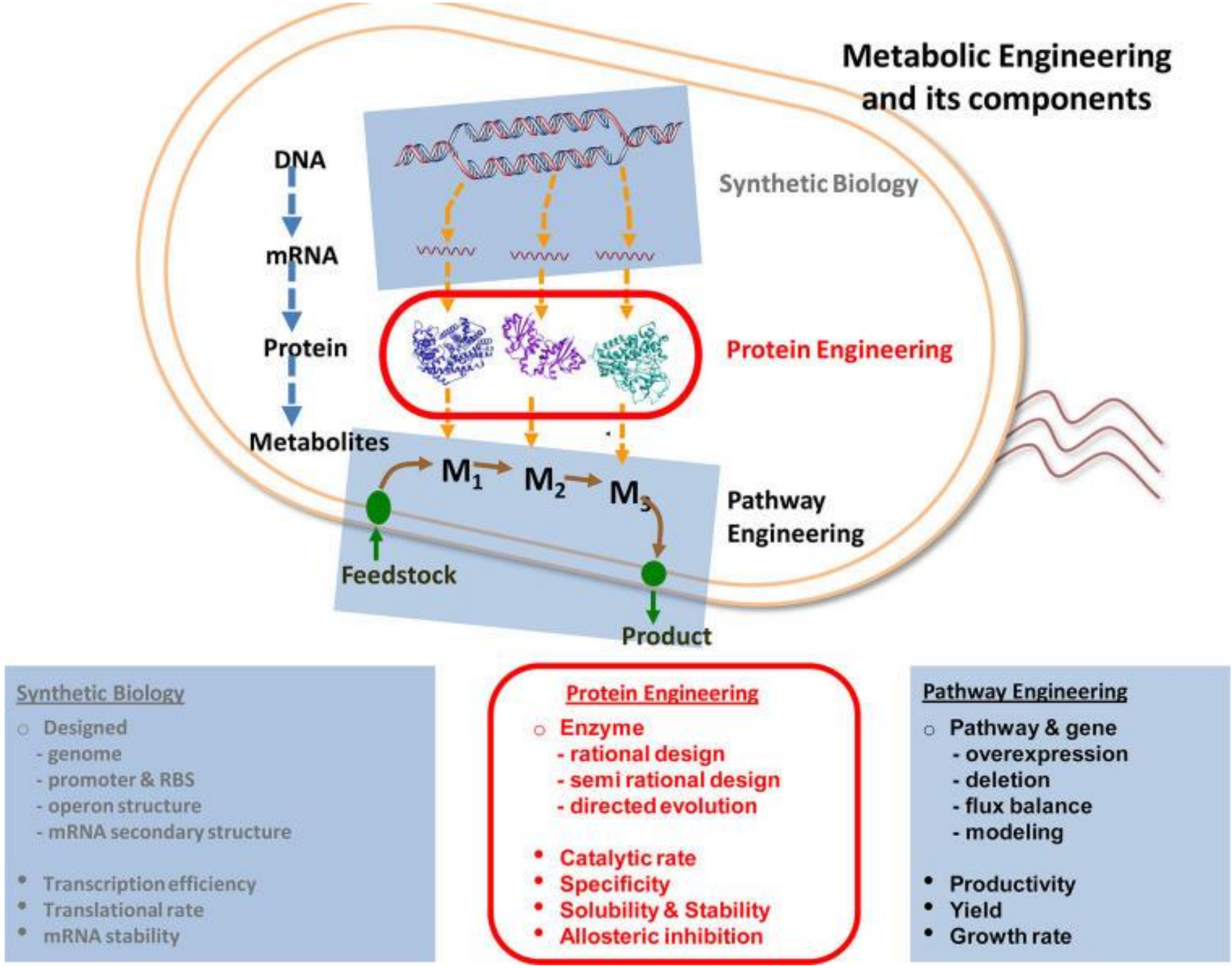


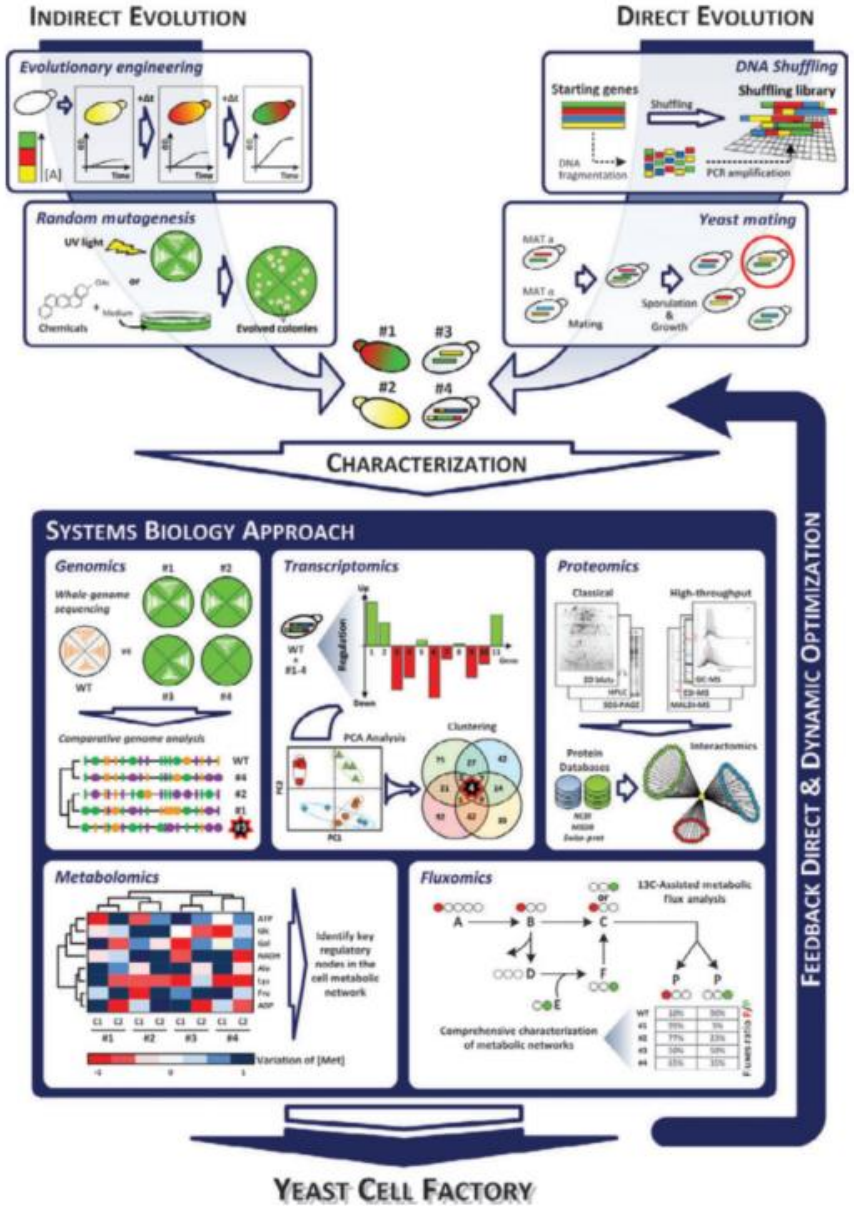
traditional approach in bioprocess design



recommended view on bioprocess design, with a reversed perspective in three dimensions. This turns value chain design into retro design, forecasting into backcasting and scale-up into scale-down

Looking at Biotechnology through the perspective of a Chemical Engineer



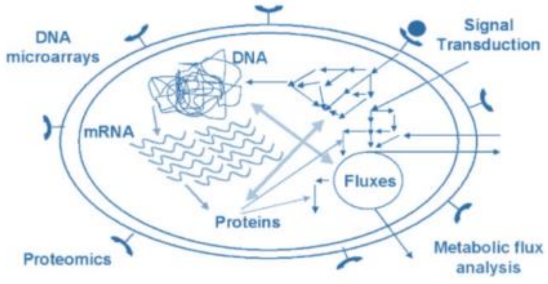


Stephanopoulos, G. (2012). "Synthetic Biology and Metabolic Engineering." ACS Synthetic Biology 1(11): 514-525.

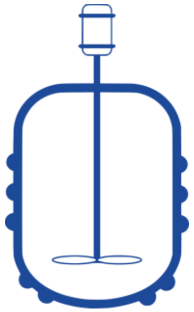
Kim, I.-K., A. Roldão, V. Siewers and J. Nielsen (2012). "A systems-level approach for metabolic engineering of yeast cell factories." FEMS Yeast Research **12**(2): 228-248.

“Chemical engineers conceive and rigorously solve problems on a ‘**continuum’ of scales**’ ranging from micoscale to macroscale”

Microscale
($\leq 10^{-3}$ m)



Mesoscale
(10^{-3} - 10^{2} m)

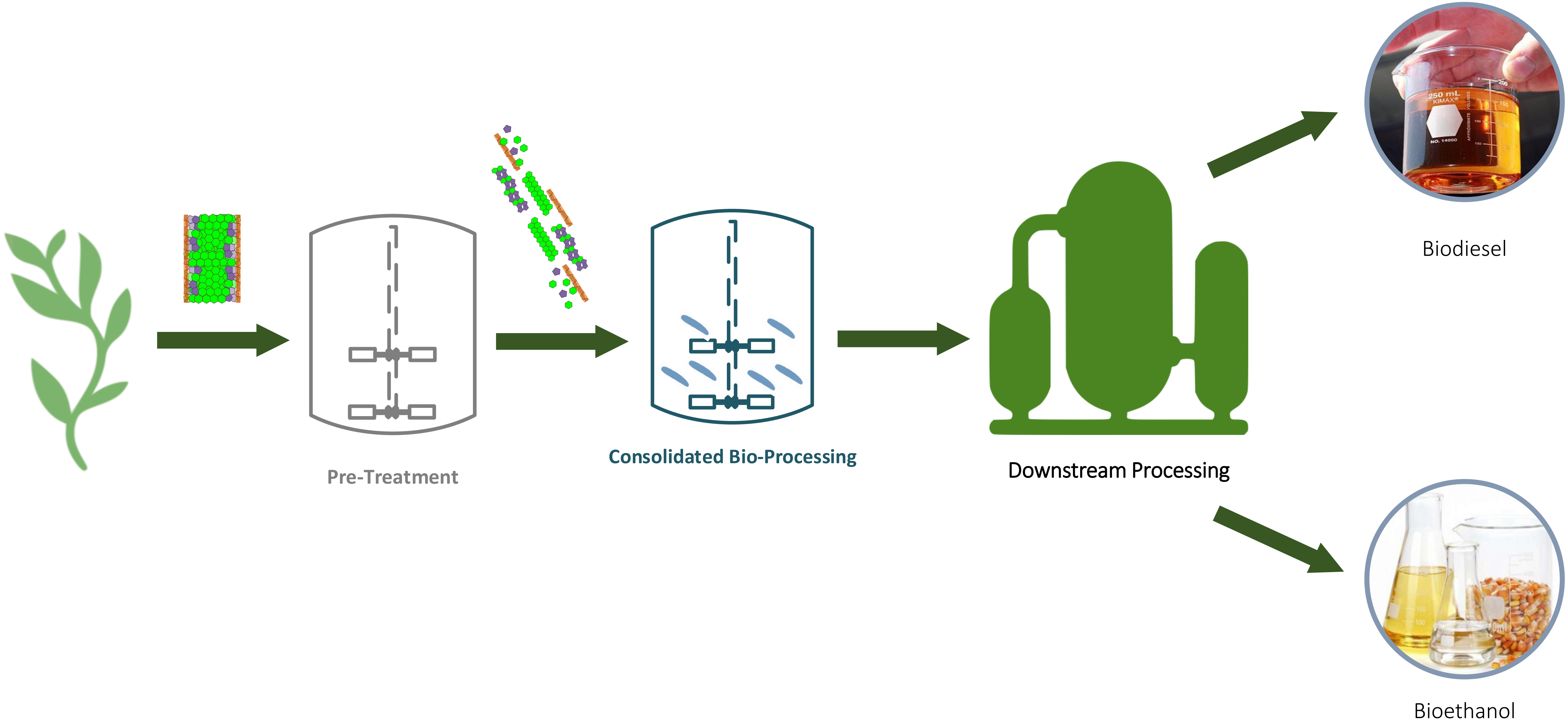


Macroscale
(> 10^{2} m)

CHEMICAL ENGINEERING SCIENCES

National Research Council (1988). Frontiers in Chemical Engineering: Research Needs and Opportunities, National Academies Press.

On addressing our country’s need for biofuels



As a research group we see that we are…

- A community of researchers bridging knowledge gaps and developing technologies on biomass utilization and biofuel production.
- A relevant research group influencing national policies and laws facilitating the development of a profitable industry based on biomass utilization and biofuel production.

BIOMASS B&B BIOFUEL ENGINEERING Research and Development GROUP

As a research group, we…

- Promote education and research towards an effective and efficient use of local biomass resources for a sustainable bio-based industry.
- Provide scientifically sound, innovative alternatives and solutions in biomass processing for the benefit of the local, national, and global communities.

Thank you for your attention!

Any comments or questions?